#7

REISSUE LITIGATION

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

In re Reissue Application of: U.S. Patent No. 5,570,338 Mark L. Collins *et al.*	Group Art Unit: 1634 Examiner: *[Unassigned]* Johannsen
Reissue Serial No. 09/533,906	Atty. Docket No. Collins Reissue Protest
Reissue Application Filed: March 8, 2000	
For: TARGET AND BACKGROUND CAPTURE METHODS WITH AMPLIFICATION FOR AFFINITY ASSAYS	

PROTEST UNDER 37 C.F.R. § 1.291(a)

TRANSMITTAL LETTER

Attn: **REISSUE LITIGATION BOX 7**
Commissioner for Patents
Washington, D.C. 20231

Sir:

Attached please find the following documents pertaining to the above-referenced Reissue Litigation:

1. Protest Under 37 C.F.R. §1.291(a) - 37 pgs.;
2. Attachment A - '338 Patent History with Post-Issuance Correction & Amendments - 1 pg.
3. Declaration of Michael M. Harpold, Ph.D. - 18 pgs.;
4. Exhibit A -Curriculum Vitae of Michael M. Harpold, Ph.D. - 9 pgs.;
5. Information Disclosure Statement Accompanying Protest Under 37 CF.R. §1.291(a) -5 pgs.
6. PTO Form 1449 - 2 pgs. (in duplicate - 4 pgs. total);
7. Twenty-One (21) References;
8. Proof of Service to: Jean Burke Fordis of Finnegan, Henderson, Farabow, Garrett, & Dunner, LLP, 1300 I Street, N.W., Washington, D.C. 20005 - 2 pgs.; and
9. Return/Prepaid Postcard

CERTIFICATE OF DELIVERY

I hereby certify that this correspondence (along with any referred to as being attached or enclosed) is being hand delivered to Group Art Unit 1634 on the date indicated below addressed to the Commissioner for Patents, Reissue Litigation Box 7, Commissioner for Patents, Washington, D.C. 20231.

8/2/00
Date of Delivery

Thinh Nguyen
Name of Person Delivering Paper

Thinh Nguyen
Signature of Person Delivering Paper

PROTEST UNDER 37 C.F.R. § 1.291(a) | **REISSUE LITIGATION**

TRANSMITTAL LETTER

Reissue App. of U.S. Patent No. 5,570,338
Mark L. Collins *et al.*

Atty. Docket No. Collins Reissue Protest

Reissue Application Filed: March 8, 2000

No fee is believed due in connection with this filing. If, however, Applicant is in error and a fee is due, please debit Deposit Account No. 07-0835 the appropriate amount.

Respectfully submitted,

Dated: August 1, 2000

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IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

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Reissue Serial No. 09/533,906	)	Examiner: *[Unassigned]*
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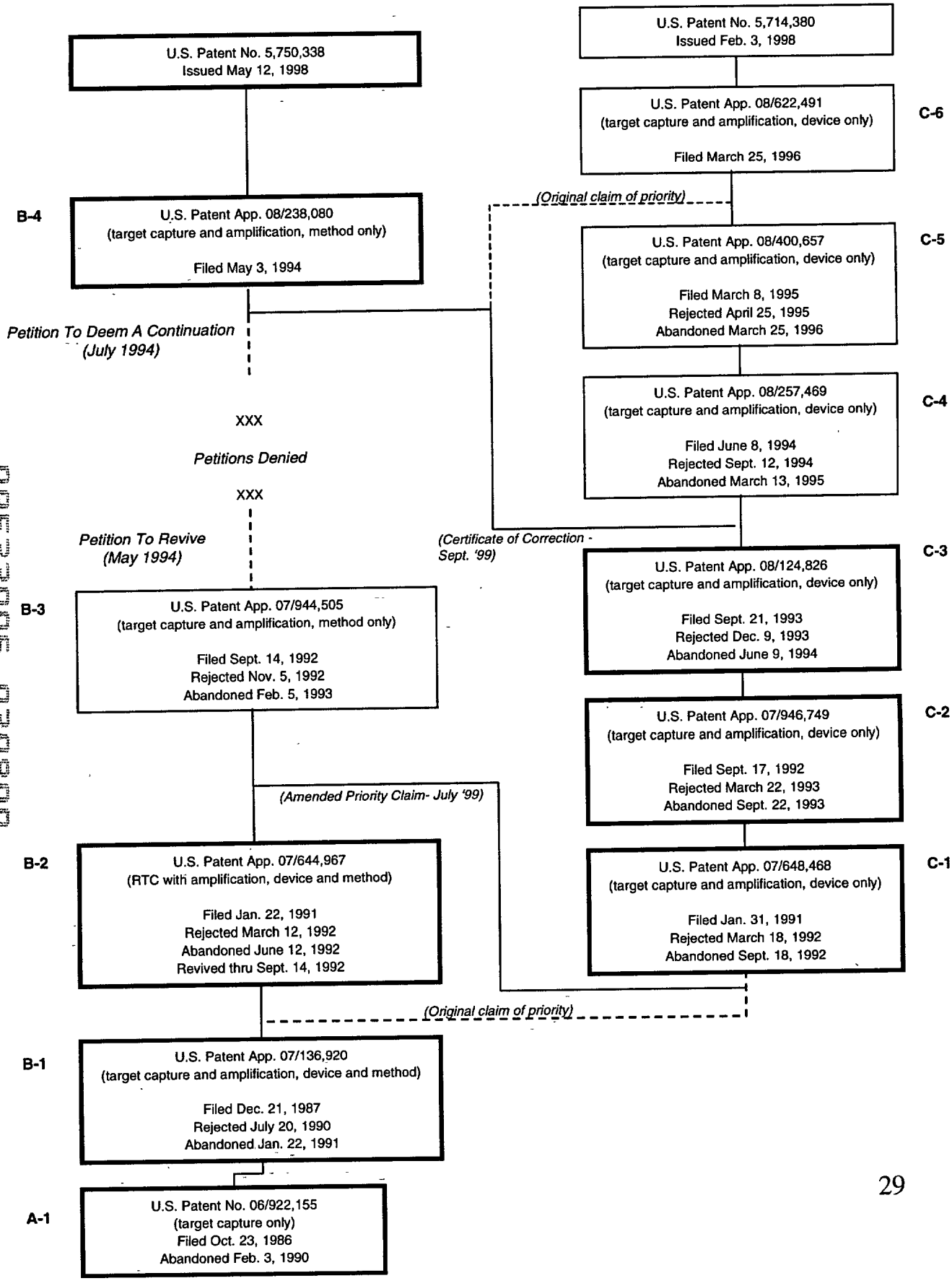
ATTACHMENT A

'338 Patent History with Post-Issuance Correction and Amendments

'338 PATENT HISTORY WITH POST-ISSUANCE CORRECTIONS AND AMENDMENTS



REISSUE LITIGATION

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

In re Reissue Application of:
U.S. Patent No. 5,750,338

Mark L. Collins *et al.*

Reissue Serial No. 09/533,906

Reissue Application Filed: March 8, 2000

For: TARGET AND BACKGROUND CAPTURE METHODS WITH AMPLIFICATION FOR AFFINITY ASSAYS

Group Art Unit: 1634

Examiner: Unassigned

PROTEST UNDER 37 C.F.R. § 1.291

ATTENTION: REISSUE LITIGATION BOX 7
Assistant Commissioner for Patents
Washington, D.C. 20231

Sir:

In accordance with 37 C.F.R. § 1.291, the following protest is submitted for consideration. This protest has been served on the reissue applicants in accordance with 37 C.F.R. § 1.248, as indicated by the attached proof of service.

CERTIFICATE OF DELIVERY

I hereby certify that this paper (along with any referred to as being attached or enclosed) is being hand delivered to Group Art Unit 1634 on the date shown below, addressed to the Commissioner for Patents, Reissue Litigation Box 7, Washington, D.C. 20231.

Thinh Nguyen
Name of Person Delivering Paper

8/2/00
Date of Delivery

Thinh Nguyen
Signature of Person Delivering Paper

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REMARKS

The following remarks present arguments supporting the conclusion that there is no basis for reissue of U.S. Patent No. 5,750,338 because:

- All claims are invalid as obvious under 35 U.S.C. § 103 (Sections I, II);
- Many claims are invalid as anticipated under 35 U.S.C. § 102 (Section III); and
- The new claims added to the reissue application should be rejected under the "written description" requirement of 35 U.S.C. § 112, first paragraph and the requirement of 35 U.S.C. § 251 that the reissue application be for the "invention disclosed in the original patent" (Section IV).

Protestor respectfully requests that the Examiner consider these remarks, and the accompanying declaration of Dr. Michael Harpold in support of the remarks contained in Sections I, II, and III, in examining reissue application no. 09/533,906.

I. ALL CLAIMS OF THE REISSUE APPLICATION ARE OBVIOUS OVER PRIOR ART ANTEDATING DECEMBER 21, 1987.

The reissue application under protest contains 59 claims. Claims 1-40 are essentially unchanged from the correspondingly numbered claims of the '338 patent. New claims 41-59 have been added in this reissue application. All the claims are directed to a process (or a kit for carrying out such process) for amplifying and/or detecting a target polynucleotide contained in a sample.

The process claimed in the reissue application involves three essential steps: contacting the sample with a solid support that binds the target polynucleotide; separating the support and bound target polynucleotide from the sample; and amplifying the target polynucleotide. These

three steps characterize the claimed invention, although additional steps and limitations are added in various claims. Claims are also directed to kits comprising means for carrying out these steps.

Protestor sets forth below reasons why the claimed invention would have been obvious prior to the filing date. Protestor also submits herewith the declaration of Dr. Michael Harpold, which supports these remarks. Dr. Harpold's declaration sets forth the general state of the art prior to the filing date (¶ 5); his view of the level of ordinary skill in the art of molecular biology at the filing date (¶ 6); a discussion of specific references relevant to the claimed invention (¶¶ 8-10); his conclusion that the claimed invention would have been obvious to one of ordinary skill in the art (¶ 11); and his observations on the declaration of Dr. David Persing, which was submitted by reissue applicants during the prosecution of the '338 patent (¶¶ 12, 13).

Each of the steps recited in the claimed method is disclosed in the prior art. Contrary to representations made by the reissue applicants during prosecution of the '338 patent, the prior art provided ample motivation to one skilled in the art at the filing date[1] to combine these steps. Consequently, the claims are unpatentable under 35 U.S.C. §103.

[1] For purposes of this section, Protestor has relied on prior art having effective dates earlier than December 21, 1987, the filing date of U.S. application no. 136,920. Although the '338 patent recites an earliest priority date of October 23, 1986, *i.e.* the filing date of U.S. application no. 922,155, the claims in the reissue application cannot be entitled to that date since the '155 application contains no disclosure whatsoever of amplification following target capture. Reissue applicants have implicitly acknowledged that they are not entitled to the priority of the '155 application (see, p.6 of the Preliminary Amendment filed with the reissue application, where applicants treat December, 1987 as the relevant prior art date).

A. Isolation Of Target Polynucleotide From A Sample By Capture On A Solid Support Is Disclosed In The Prior Art.

U.S. Patent No. 4,672,040 (*Josephson*) discloses contacting capture probes immobilized on dispersible magnetic beads with a sample containing complementary target polynucleotides and separating the support and bound polynucleotides from the target, *i.e.*, the first two steps of the presently claimed process. In particular, *Josephson* states:

> Specific DNA or RNA fragments can also be isolated from genomic and cloned DNA by immobilization of a known probe to the magnetic particles and placing the coupled particles in contact with a mixture of nucleic acid fragments, including the desired species. After hybridization the particles may be magnetically separated from unbound materials, washed, and the hybridized molecules isolated. (Col. 19, ll. 3-10)

The foregoing statement directly contradicts the assertion in the '338 patent that, "...[M]agnetic particles have not been suggested as retrievable supports for target capture and background removal" (col. 4, ll. 27-29). *Josephson* further states that the magnetic particles can be used in binding assays (col. 16, ll. 13 *et seq.*). Clearly, such binding assays include nucleic acid hybridization assays since the use of dispersible capture probes for nucleic acid hybridization is taught, for example, in Section 6.6 (col. 18, ll. 29 *et seq.*).

U.S. Patent No. 4,554,088 (*Whitehead*) discloses the use of single-stranded nucleic acid bound to dispersible magnetic beads to isolate complementary nucleic acid from a sample. The nucleic acid capture probe immobilized on a magnetic bead is employed as a "ligand" to bind its soluble "ligate", which is the complementary nucleic acid in the sample (see Table III, col. 17). The magnetic beads bound to target nucleic acid are magnetically separated from the sample and unbound (non-target) species are removed by washing (col. 17, ll. 36-40).

Polsky-Cynkin et al., Clin. Chem. 31/9:1438-1443(1985) describe DNA hybridization assays in which target DNA is captured by a complementary probe affixed to a solid support. The captured target DNA on the solid support is separated from the sample and detected with a radiolabeled probe. The solid supports employed included agarose beads (*i.e.,* retrievable supports), polypropylene test tubes and polypropylene solid-phase receptacles (*e.g.*, see p. 1439).

U.S. Patent No. 4,563,419 (*Ranki*) discloses a hybridization assay in which target nucleic acid is isolated from a sample by hybridizing it to a complementary capture probe immobilized on a solid support (nitrocellulose filter) and separated from the sample by washing. The captured target polynucleotide is detected with a labeled probe.

B. Amplification Of Target Polynucleotides Is Disclosed In The Prior Art.

The reissue application defines the term "amplify" in extremely broad terms. The definition includes any process by which copies of target polynucleotide are produced or by which other molecules (sometimes referred to as "reporter" molecules) are produced by virtue of the presence of the target polynucleotide (col. 2, ll. 9-19).

The applicants' broad definition encompasses processes such as cloning, cell-free translation and synthesis of cDNA from mRNA, all of which were basic techniques of molecular biology in December 1987. The Examiner will appreciate that many references describing such forms of amplification could be cited as prior art. New claims 41-59, which were added in the reissue application, limit the amplification step to an *in vitro* amplification process in an attempt

to avoid what reissue applicants characterize as "prior art from an earlier period of molecular biology"[2] (Preliminary Amendment, p.5). Consequently, here Protestor will focus on prior art references disclosing *in vitro* amplification.

The specification of the '338 patent discloses only three types of *in vitro* amplification: non-specific enzymatic amplification primed by random hexamer primers (Examples 5 and 6); non-specific amplification using Qß replicase enzyme (Example 7); and non-specific transcription of DNA by *E. coli* RNA polymerase (Example 4). To the extent that any of these might be effective to replicate target polynucleotide, they are each disclosed in the prior art. The use of random hexamer oligonucleotide primers to initiate non-specific enzymatic reproduction of polynucleotides is disclosed in *Feinberg et al., Anal. Biochem. 132:6-13 (1983).*

Example 7 of the '338 patent purports to disclose non-specific, exponential amplification of isolated polynucleotide using the enzyme Qß replicase. To the extent that the reissue applicants rely on the described Qß replicase amplification to support the claims, they acknowledge, at col. 32, ll. 16-17, that this form of amplification is taught in the prior art (*Blumenthal, Proc. Natl. Acad. Sci. U.SA. 77:2601 (1980)).*

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[2] Cell-free translation and cDNA synthesis are both *in vitro* processes, as are the initial steps of many cloning procedures. Therefore, the limitation in new claims 41-59 to *in vitro* amplification does not avoid the effect of prior art references teaching such methods. Having chosen to define "amplify" broadly, applicants cannot now seek a narrower definition in an attempt to avoid the prior art. That is, the patentee cannot now offer an interpretation that would alter the record (the specification) on which the public is entitled to rely. *Vitronics Corp. v. Conceptronic Inc.*, 90 F.3d 1576, 1582-1583, 39 USPQ2d 1573, 1577 (Fed. Cir. 1996).

The use of *E. coli* RNA polymerase core enzyme to transcribe DNA into RNA *in vitro* was known in the prior art, as acknowledged by applicants in their citation to R. Burgess in *RNA Polymerase, Cold Spring Harbor Press, pp. 69-100 (1976)* ('338 patent, col. 30, ll. 62-64).

In addition to the three forms of non-specific *in vitro* amplification disclosed in the '338 patent specification, the prior art discloses other forms of *in vitro* amplification. The well-known polymerase chain reaction (PCR), which is an *in vitro* exponential amplification method, is disclosed, for example, in *U.S. Patent No. 4,683,202 (Mullis)*. A method of amplifying RNA sequences by strand displacement synthesis is disclosed in *Gaubatz et al., Biochim. Biophys. Acta, 825*:175-187 (1985)[3].

C. The Prior Art Provides Motivation To Combine Target Capture On A Solid Support With Amplification Of The Isolated Polynucleotide.

Contrary to arguments made during prosecution of the '338 patent, the prior art explicitly suggests combining the isolation of a target polynucleotide from a sample by capture on a solid support with subsequent amplification of the isolated polynucleotide.

In *Methods of Gene Isolation (Brown et al., Ann. Rev. Biochem., 43:667-693 (1974))*, the authors review various methods for isolating nucleic acid sequences of interest for analysis, *e.g.* by molecular hybridization assay. The authors describe different methods of purifying

[3] See Section III for a more detailed discussion of this reference.

polynucleotides of interest from samples. In the section entitled "Polynucleotides Fixed to Insoluble Matrices" they state:

> ssDNA components can be purified by fixing complementary RNA or DNA molecules to some kind of insoluble support and circulating the soluble DNA mixture through the affinity column. Large amounts of sequence-specific DNA have been purified from mixtures of phage and bacterial DNAs by circulating the denatured DNAs through columns containing one of the DNAs adsorbed to nitrocellulose. (p. 673-674)

In their "CONCLUDING REMARKS," the authors teach the desirability of combining target isolation with a subsequent amplification step, particularly where the target polynucleotide is present in the initial sample at low concentration.

> **Clearly, purification of important structural genes will have to be coupled with some method in which a small amount of a given gene can be increased enormously in amount.** After purification has enriched the gene sequence about a thousandfold the remaining DNA would be amplified hundreds- to thousandsfold in amount...**The amplification step might be carried out in vitro by an efficient DNA polymerase, which would replicate faithfully each molecule of DNA many times.** (p. 687) (emphasis added)

The description of the amplification step in the '338 patent is a virtual echo of the emphasized passage above:

> In the situation where the target is a polynucleotide, additional target, or target-like molecules, or molecules subject to detecting **can be made enzymatically with DNA or RNA polymerases or transcriptases**. (col. 2, ll. 16-19) (emphasis added)

Thus, *Brown et al.* provide a clear direction to combine solid phase target capture with amplification to provide large quantities of target polynucleotide for detection. This goes to the very core of the reissue applicants' claimed "invention" because during prosecution applicants' attorney stated, "The invention advances nucleic acid hybridization methods by combining target purification methods with target amplification methods." (Preliminary Amendment and Response to Restriction Requirement, filed December 5, 1995 in U.S. application no. 08/283,080 ("the '080 application")).

The reissue applicants cannot effectively argue that the *Brown et al.* article may not have provided all the enabling technical details for carrying out the steps of the claimed process. Both solid phase target capture and amplification methods (as described in the '338 patent) were well known to those skilled in the art by December 21, 1987, as shown by the references cited in Sections A and B above.

Additional motivation to combine target capture on a solid support with amplification is provided by *Arsenyan et al., Gene 11:97-108 (1980)*[4]. This reference describes the isolation and amplification of rat liver 5S RNA genes. The authors state:

> In order to study the arrangement of genes it is necessary to isolate **amplified** homogeneous DNA sequences with spacer regions. There are two principal ways of isolating such sequences: (a) "Shotgun" cloning of total genomic DNA, followed by a colony hybridization with labelled RNA or cDNA...Since most of the eukaryotic genes are found as single copies in a genome, the screening of individual recombinants is difficult. (b) **A preliminary enrichment of such genes, followed by a bacterial cloning.** (p. 97, col.2 to p.98, col.1, emphasis added)

[4] See Section III for additional discussion of this reference.

This reference shows that the art understood that when additional sensitivity and specificity were needed, the combination of preliminary enrichment and amplification would yield better results. That is, if genes were present in multiple copies, then shotgun cloning would suffice. However, if single copy genes were sought, then shotgun cloning was insufficient, and it would be necessary to combine preliminary enrichment with an amplification process.

Arsenyan et al. accomplished the preliminary enrichment of the 5S RNA gene by capture of the individual (+) and (-) strands from a sample containing denatured rat liver DNA fragments on solid supports (oligo(dT) cellulose or 5S cDNA-cellulose) having complementary capture probes bound thereto. The strands were separated from the sample, annealed, cloned into *E. coli* and amplified by growing the transformants.

Arsenyan et al. provide express motivation to purify target DNA from a sample by capture on a solid support as a preliminary step to amplification. Moreover, and significantly, *Arsenyan et al.* specifically refer to the product of the cloning step as "amplified" DNA. Thus, the motivation is provided to combine target capture with any form of amplification, including *in vitro* amplification. This is consistent with reissue applicants' own definition of amplification (col. 2, ll. 9-19), which equates all forms of amplification, provided only that they produce additional target molecules or target-like molecules.

Syvanen et al., Nucleic Acids Res., 14(12):5038-5048 (1986) provide further motivation to combine target capture with target amplification by stating that "the sensitivity of the [sandwich hybridization] method can be increased ... by amplifying the target DNA" and referring to a reference that describes PCR amplification. (p. 5044, ll. 11-13.)

D. Summary And Application Of The Prior Art To The Claims.

In view of the foregoing discussion of the prior art, Protestor submits that claims 1-59 of the reissue application are unpatentable under 35 U.S.C. §103 because they are obvious over *Josephson*, *Whitehead*, *Polsky-Cynkin et al.* or *Ranki* taken with *Feinberg et al.*, *Blumenthal*, *Mullis* or *Gaubatz et al.*, in view of *Brown et al.*, *Arsenyan et al.* or *Syvanen et al.* Each of *Josephson*, *Whitehead*, *Polsky-Cynkin et al.* and *Ranki* disclose the capture of target polynucleotides on solid supports and separation of the support and bound target from the sample. Each of *Feinberg et al.*, *Blumenthal*, *Mullis* and *Gaubatz et al.* disclose *in vitro* amplification methods. *Brown et al.* provide motivation to combine target capture on solid supports with amplification. *Arsenyan et al.* and *Syvanen et al.* provide additional motivation to combine target capture on solid supports with amplification.

Claim 1 is made obvious by a combination of these references that includes any one of the cited target capture references and any one of the cited *in vitro* amplification references, in view of the motivation provided by *Brown et al.* and/or *Arsenyan et al.* to combine these steps.

Claims 2 and 8 recite a "retrievable" support. Both *Josephson* and *Whitehead* disclose supports that are retrievable. *Polsky-Cynkin et al.* disclose the use of a retrievable support in a hybridization assay that includes the detection step of claim 8.

Claims 3 and 9 recite the inclusion of a probe on the solid support, which is also disclosed in *Josephson*, *Whitehead*, *Polsky-Cynkin et al.* and *Ranki et al.*

Claims 4-6, 10-12, 17, 18, 29-33, 35-37, 39 and 40 are dependent claims that recite the use of various polymerases to amplify the target. The use of such polymerases to amplify target sequences is disclosed in *Feinberg et al.*, *Blumenthal*, *Mullis* and *Gaubatz et al.*

Claim 7 is directed to a method of detecting the target polynucleotide and recites the additional step of detection. *Polsky-Cynkin et al.* and *Ranki* disclose hybridization assays that involve a detection step. Moreover, *Josephson* and *Whitehead* both indicate that the target capture particles disclosed therein can be used in binding assays. These would necessarily include a detection step.

Claims 13, 14 and 16 add the limitation that the amplified polynucleotide is contacted with a label or labeled probe, which is obvious in view of *Polsky-Cynkin et al.*, who disclose a radiolabeled probe (p. 1439, col. 2); *Ranki* who discloses a labeled probe (col. 6, ll. 10-35; col. 7, ll. 20-23 and ll. 35-40) for detection in an assay; or *Josephson*, who discloses a variety of labels (col. 15, ll. 52-58).

Claim 19 is directed to a method of detection that recites the additional step of contacting the amplified target with a second solid support and a detection probe. Claim 15, which is dependent on claim 7, contains essentially the same limitation. The use of a second capture probe to separate the amplified target and detection probe from the other components of the amplification reaction would be obvious in view of *Josephson*, *Whitehead* and *Ranki.*

Claims 20-26 are directed to "kits" for carrying out the claimed methods. Since the methods are obvious for reasons previously stated and the reagents for carrying out the methods

are well known, the mere assemblage of those reagents into a "kit" for carrying out the method cannot confer patentability.

Independent claims 27, 28, 34 and 38 are directed to methods of amplification or detection that recite the use of a nucleic acid probe that is capable of binding to the target nucleic acid as well as to the solid support, thereby providing the means for binding the target to the support. In claims 27 and 28, the probe is first allowed to hybridize to the target before contacting it with the solid support. In claims 34 and 38, the probe, target and solid support are brought into contact in a single step. These claims are all obvious over the references cited above in connection with claim 1, further in view of *Syvanen et al.* Each of *Josephson, Whitehead, Polsky-Cynkin et al.* and *Ranki* disclose the use of a nucleic acid probe that binds the target polynucleotide and the solid support. *Syvanen et al.* teach the advantage of allowing the interaction of the capture probe and target to occur in solution. *Syvanen et al.* disclose a hybridization assay in which the target nucleic acid is captured on a solid support (agarose beads) by the use of a capture probe capable of hybridizing to the target and binding the agarose beads through a biotin-avidin interaction. The capture probe and target are allowed to hybridize in solution prior to capture on the solid support. The advantages of allowing the capture probe to interact with the target in solution are specifically pointed out at pages 5042-5043 ("Kinetics of the reaction"). *Arsenyan et al.* disclose a probe (poly-A tailed 5S rRNA) that binds to both the target polynucleotide (5S DNA) and the support (oligo-dT) by nucleic acid hybridization.

Claims 41-59 are new claims presented for the purpose of reissue. All are dependent on claims in the issued '338 patent.

Claims 41, 47, 53 and 56-59 recite that "the target polynucleotide is amplified *in vitro* to produce a multitude of amplification products." Similarly, claims 54 and 55, which are dependent upon kit claims 20 and 24, recite that the "means for amplifying provide for *in vitro* amplification of the target polynucleotide to produce a multitude of polynucleotide amplification products." These limitations cannot confer patentability on otherwise unpatentable claims, since *Feinberg et al.*, *Mullis* and *Gaubatz et al.* disclose *in vitro* amplification methods that produce a multitude of amplification products.

Claims 42, 45, 48 and 51 recite that the *in vitro* amplification is linear or exponential. Claims 43 and 49 further limit the amplification to exponential amplification. *Feinberg et al.* discloses *in vitro* amplification that is linear. *Gaubatz et al.* indicate (p. 180, col. 1) that their strand displacement method of reproducing polynucleotides exhibits an exponential phase. *Mullis* discloses *in vitro* amplification methods that are exponential. While Protestor questions whether the use of Qß replicase enables *in vitro* exponential amplification as described in the '338 disclosure (Example 7), to the extent that reissue applicants rely on this method to support such amplification, it is disclosed in *Blumenthal.*

Claims 44 and 50 recite that the target polynucleotide is amplified with a polymerase and at least one oligonucleotide primer. *Feinberg et al.*, *Mullis* and *Gaubatz et al.* each disclose amplification using a polymerase and at least one oligonucleotide primer.

Claims 46 and 52 each recite the use of more than one polymerase to perform amplification. This limitation is made obvious by the *Gaubatz et al.* disclosure that uses a reverse transcriptase to synthesize first-strand cDNA followed by DNA polymerase I (Klenow) to synthesize second strand cDNA (p. 176, col. 2) and for strand displacement (Fig. 1).

In summary, based on the foregoing discussion, Protestor submits that:

- Claims 1, 7, 19, 20 and 24 are obvious over any one of *Josephson*, *Whitehead*, *Polsky-Cynkin et al.* or *Ranki* combined with any one of *Feinberg et al.*, *Blumenthal*, *Mullis* or *Gaubatz et al.*, in view of *Brown et al.* or *Arsenyan et al.* Dependent claims 2-5, 8-11, 13-18, 21, 22, 25, 41, 42, 45, 47, 48, 51, 53, 54, and 55 are obvious over the same references.
- Dependent claims 6, 12, 23, 26, 44, and 50 are obvious over any one of *Josephson*, *Whitehead*, *Polsky-Cynkin et al.* or *Ranki* combined with any one of *Feinberg et al.*, *Mullis* or *Gaubatz et al.*, in view of *Brown et al.* or *Arsenyan et al.*
- Dependent claims 43 and 49 are obvious over any one of *Josephson*, *Whitehead*, *Polsky-Cynkin et al.* or *Ranki* combined with *Mullis* or *Gaubatz et al.*, in view of *Brown et al.* or *Arsenyan et al.*
- Dependent claims 46 and 52 are obvious over any one of *Josephson*, *Whitehead*, *Polsky-Cynkin et al.* or *Ranki* combined with *Gaubatz et al.*, in view of *Brown et al.* or *Arsenyan et al.*

- Claims 27, 28, 34 and 38 are obvious over any one of *Josephson*, *Whitehead*, *Polsky-Cynkin et al.* or *Ranki* combined with any one of *Feinberg et al.*, *Blumenthal*, *Mullis* or *Gaubatz et al.*, in view of *Brown et al.*, *Arsenyan et al.* or *Syvanen et al.* Dependent claims 29-30, 32, 35, 36, 39 and 56-59 are obvious over the same references.

- Dependent claims 31, 33, 37, and 40 are obvious over any one of *Josephson*, *Whitehead*, *Polsky-Cynkin et al.* or *Ranki* combined with any one of *Feinberg et al.*, *Mullis* or *Gaubatz et al.*, in view of *Brown et al.*, *Arsenyan et al.* or *Syvanen et al.*

II. ARGUMENTS MADE BY THE APPLICANTS IN THE PROSECUTION OF THE '338 PATENT CANNOT OVERCOME AN OBVIOUSNESS REJECTION.

In January 1997, the Examiner rejected the claims of the '080 application over a combination of references showing solid phase target capture with references showing amplification by PCR. In response, the applicants put forth an essentially two-pronged reply. First, applicants argued that the prior art provided no expressed motivation to combine the teachings of target capture and PCR. See, *e.g.*, Examiner's Interview Summary, paper no. 14. Second, applicants sought to focus discussion on PCR amplification, asserting that their invention solved previously unrecognized problems with PCR and represented an improvement of the PCR process. Neither of applicants' arguments is sufficient to overcome an obviousness rejection.

Applicants' first argument that the prior art provided no motivation to combine target capture and amplification is clearly refuted by the *Brown et al.* and *Arsenyan et al.* references, discussed above, which expressly suggest such combination. Moreover, the suggestions in *Brown et al.* and *Arsenyan et al.* to combine target capture with amplification render moot applicants' second argument regarding the asserted solution of unsolved problems with PCR amplification and alleged improvements in PCR amplification. *Brown et al.* suggest that small quantities of target polynucleotide can be extracted from a sample by sequence-specific target capture on a solid support and then amplified *in vitro* to produce sufficient quantities for further analysis. As long as some motivation or suggestion to combine prior art references is found in the prior art, the law does not require that the prior art suggest combining them for the same reasons set forth by the inventor. *In re Beattie*, 974 F.2d 1309, 1312, 24 USPQ 2d 1040, 1042 (Fed. Cir. 1992); *In re Kronig*, 539 F.2d 1300, 1304, 190 USPQ 425, 427-428 (CCPA 1976); *In re Lintner*, 458 F.2d 1013, 1016, 173 USPQ 560, 562 (CCPA 1972).

The declaration of Dr. David Persing, filed immediately before allowance of the '338 patent, asserted that applicants had solved unrecognized problems with PCR and improved the PCR process. The reissue applicants' arguments for patentability were so focused on PCR that the Examiner was moved, in her statement of reasons for allowance, to state that "*The claims are drawn to methods of PCR amplification* wherein the target is first separated from the sample by using a support that binds to the target polynucleotide and then amplified." ('080 application, Examiner's Interview Summary, paper 22, emphasis added).

In fact, PCR is neither disclosed in the specification nor claimed. Clearly the claims are not *limited* to PCR or to any other form of target-specific amplification. Arguments made by the reissue applicants during prosecution of the '338 patent must be scrutinized in the context of the claims *as actually presented*, not narrow claims containing a non-existent limitation to PCR.

In considering the issue of obviousness, the claims must be given their broadest possible interpretation, within the boundaries imposed by the written description requirement. *In re Baker Hughes Inc.*, ___ F.3d ___, 55 USPQ2d 1149, 1152 (Fed. Cir. 2000); *In re Tanaka*, 551 F.2d 855, 859, 193 USPQ 138 (CCPA 1977). Narrow arguments directed to alleged improvements to PCR, in particular, or the solution of unexpected problems with PCR are simply not relevant to consideration of the obviousness references set forth above. To allow claims based on PCR-focused arguments would effectively permit the applicants to base patentability on a limitation that is not present in any claim and is not even disclosed in the specification. Therefore Dr. Persing's statements about PCR cannot overcome an obviousness rejection when the claims also encompass other amplification methods.[5]

One of the primary PCR-focused argument advanced during prosecution of the '080 application was that those skilled in the art would not have been motivated to combine target capture with *PCR* because the problem of non-specific amplification, *i.e.*, amplification of non-target sequences, was not recognized at the filing date of the application. The Persing

[5] See also Paragraph 12 of the accompanying declaration of Dr. Michael Harpold, which discusses Dr. Persing's declaration. Dr. Harpold concludes that Dr. Persing inaccurately describes the prior art and implicitly assumes the level of skill in the art at the filing date to be lower that it actually was.

declaration, filed on July 14, 1997, states:

> Initially, users and proponents of PCR believed that PCR was **highly specific** and could be made to selectively amplify the desired target in an otherwise complex sample system. Practitioners believed that adequate specificity could be imparted to the amplification by careful selection of the primers used in the amplification so that additional steps for isolating target prior to amplification were not required. (Persing Decl. ¶6) (emphasis added).[6]

Dr. Persing's statements, in contrast to the broad scope of the claims, are confined to PCR. Such limited statements are irrelevant to the obviousness of the invention. The relevant art must be considered in light of the actual claims, broadly construed -- not claims construed narrowly as if they included a limitation to PCR that they, in fact, do not include. Those skilled in the art would immediately recognize that use of non-specific amplification methods, as taught in the specification and encompassed by the claims, would result in the indiscriminate amplification of polynucleotide sequences in the sample unless some prior enrichment of the

[6] Dr. Persing's statement as quoted above is contradicted by applicants' own attorney, Norval Galloway, who acknowledged earlier in the '080 application prosecution that Mullis recognized that non-specific amplification could be a problem with PCR. In responding to a rejection over a combination of references including *Mullis U.S. Patent No. 4,683,202*, Mr. Galloway stated "At the same time, however, Mullis recognized that non-target background nucleic acids might also be amplified in addition to the intended target nucleic acids." Preliminary Amendment and Response to Restriction Requirement, filed December 5, 1995. See also, *"DNA Cleavage Adapter Groomed For Genetic Diagnostics," Biotechnology Newswatch 6 (19):8 (1986)*, in which Mullis is quoted as recognizing that, despite specific primers, PCR results in "a lot of other things replicating that you don't want" and suggesting combination of PCR with other techniques to improve specificity; and O*rkin, N. Engl. J. Med. 317(16):1023-5 (1987)*, which describes the use of elevated temperature in PCR as one approach to the problem of background amplification due to hybridization of primers to non-target sequences (p. 1024, col. 2). Thus, even if the claims were directed to an improved PCR method, the art and applicants' admission establish a recognized need that provides the motivation to combine the references.

target polynucleotide was performed. The benefit of isolating target prior to amplification would, therefore, be obvious.

Other arguments advanced by the reissue applicants during prosecution of the '080 application are also insufficient to overcome an obviousness rejection. For example, applicants argued that practitioners of hybridization assays were reluctant to use hybridization techniques to purify a target polynucleotide prior to amplification because the binding efficiency of a capture probe to its target is less than 100%. The only support offered for this conclusion is a statement from a 1993 publication that "[T]o date, there are no published studies that demonstrate efficient capture and detection of fewer than 100 target molecules, ..." (Persing decl., ¶13). This statement does not warrant the conclusion drawn by Dr. Persing with regard to the motivation of those skilled in the art at the December 1987 filing date. To the contrary, when viewed in the context of the state of the art in December 1987, this statement indicates a clear motivation to combine target capture and amplification methods. As the prior art cited in Section II.B shows, target capture on solid supports was already being used in hybridization assays by December 1987 (see, *Polsky-Cynkin et al.* and *Ranki*). If, as Dr. Persing asserts, those skilled in the art were concerned about loss of target during the capture step, this concern would provide motivation to employ known amplification procedures following target capture in order to compensate for loss of target, thereby increasing the sensitivity of the assay. Indeed, that is precisely what *Brown et al.* suggest -- using *in vitro* amplification to increase small amounts of target nucleic acid isolated by methods such as target capture on a solid support. Consequently,

the use of amplification was an obvious way to increase the sensitivity of art-known hybridization assays employing target capture.

Finally, the reissue applicants argued, during prosecution of the '080 patent, that the claimed method provided an added benefit that was unexpected before December, 1987, i.e. that separation of the target from the sample prior to amplification removed potential amplification inhibitors. In contrast, Protestor submits that the prior art recognized that there would be an inherent benefit of performing target purification before *in vitro* amplification that relied on polymerase activity and thus provided ample motivation to combine target capture with such amplification. The presence of polymerase inhibitors in biological samples has been known for many years. See, *e.g., DNA Synthesis, Kornberg, A., Freeman & Co., p.65 (1974)* and *Burgess, supra*[7]. Thus those skilled in molecular biology have long recognized the need to purify nucleic acids from samples prior to reproducing them with polymerase. See *Maniatis et al., Molecular Cloning, A Laboratory Manual, (Cold Spring Harbor Laboratory, Cold Spring Harbor, NY 1982) Chap. 6, Extraction, Purification and Analysis of mRNA From Eukaryotic Cells, pp. 187-196 and Chap. 7, Synthesis and Cloning of cDNA, pp. 213-214.* Thus, it would have been obvious to one skilled in the art that purification of target polynucleotide by any method, including the art-known target capture methods, would have the beneficial effect of removing

[7] *Kornberg,* a basic text on DNA synthesis, states that the presence of an endonuclease that creates 3 -phosphoryl termini can convert a template primer into an inhibitor that binds polymerase enzyme in an unproductive complex. *Burgess* also recognized the presence of contaminants in enzymatic reactions (pp. 86-89).

inhibiting substances. It should also be noted that the inventors of PCR recognized that the failure to purify target DNA from a sample could result in inhibition of amplification. *Saiki et al., Nature, 324: 163-6 (1986), at 164, col. 2*

For all of the reasons set forth above, applicants' arguments concerning PCR, and applicants' other arguments, cannot save the broad claims of the reissue application from an obviousness rejection.

III. CLAIMS 1-8, 10-14, 24-52, 55, 58 AND 59 ARE ANTICIPATED BY PRIOR ART ANTEDATING DECEMBER 21, 1987.

Claim 1 is anticipated by each of *Arsenyan et al., Gene 11:97-108 (1980); Gaubatz et al., Biochim. Biophys. Acta, 825:175-187 (1985); Boss et al., J. Biol. Chem., Vol. 256:12958-12961 (1981);* and *Powell et al., Cell 50:831-840 (1987).*[8]

Arsenyan et al. teach that a target polynucleotide (DNA encoding rat 5S ribosomal RNA) can be enriched using target capture prior to amplification. The negative strands of the target

[8] For purposes of this section, Protestor has relied on prior art having effective dates earlier than December 21, 1987, the filing date of U.S. application no. 136,920. However, the claims of the reissue application may also be anticipated by European Patent publication no. 0 328 829 A2 and by *Thompson et al., Clin. Chem. 35/9, 1878-1881 (1989)*, if the reissue application is not entitled to a priority date before January 31, 1991.

Although reissue applicants may assert that application no. 07/648,468 is entitled to the benefit of the filing date of U.S. application no. 07/644,967, the PTO has previously observed that the '468 application was *not* entitled to the benefit of the filing date of the '967 application (or any earlier application). (Paper No. 11, dated April 25, 1995, at pages 2-3, of application 08/400,657; see also 37 C.F.R. § 1.78(a)(1); *In re Chu*, 66 F.3d 292, 297, 36 USPQ2d 1089, 1093 (Fed. Cir. 1995).) Protestor believes that the issue of the priority date may not be amenable to resolution in this *ex parte* proceeding, because it may require discovery of documents and witnesses that is only available in an *inter partes* proceeding.

DNA were captured by passing them directly over a cellulose column having 5S cDNA covalently bound thereto as a target capture probe (pg. 100, col. 2, para. 2). The positive strands of the target DNA were first hybridized to poly(A)-tailed 5S RNA in solution and this hybrid complex was then captured via hybridization to an oligo(dT)-cellulose column (pg. 100, col. 2, para. 3). The columns were washed to separate the target polynucleotides from the starting material. Following target capture, the eluted positive and negative strands were re-natured, ligated into a cloning vector and transformed into *E. coli*. Growing the transformed *E. coli* necessarily produced a multitude of copies of the target polynucleotide. This multiplication of the target polynucleotide clearly falls within reissue applicants' definition of amplification (col. 2, ll. 9-19). Thus, each of the steps of claim 1 -- capture of the target on a solid support, separation from the sample and amplification -- are disclosed in this reference.[9]

Gaubatz et al. purified globin mRNA (the target polynucleotide) by chromatography on oligo(dT)-cellulose (the solid support that binds to the target polynucleotide). The purified mRNA was then converted to double-stranded cDNA containing a hairpin loop using AMV reverse transcriptase to synthesize the first strand and DNA polymerase I (Klenow) to synthesize the second strand. A poly(dC) tail was then added to the 3' end of the cDNA using terminal

[9] The reissue applicants imply, at page 12 of the preliminary amendment filed with the reissue application, that it is merely the suggestion of a licensee that the meaning of amplification in the '338 patent includes *in vivo* amplification and cell-free translation. However, the reissue applicants chose this definition in their specification (col. 2, ll. 9-19). In light of the public's right to rely on the existing specification, applicants cannot now avoid the prior art by attempting to imply a narrower definition of that term in the claims. See discussion, *infra*, at footnote 2.

deoxynucleotidyltransferase. The resulting double-stranded cDNA with a single-stranded poly(dC) tail was used as a template for amplification. An oligo$(dG)_{12-18}$ annealed to the poly(dC) tail was used to prime strand displacement synthesis, producing an inverted repeat sequence of the double-stranded cDNA. The template was "replicated many times" (see p. 179, col. 2) and the amplification/replication had an exponential phase (see p. 180, col. 3). Thus, Gaubatz *et al.* disclose each of the steps of claim 1.

Boss et al., J. Biol. Chem., 256:12958-12961 (1981) describes the isolation of yeast iso-1-cytochrome c (CYC1) mRNA (the target polynucleotide) by hybridization to cloned CYC1 DNA attached to diazobenzyloxymethyl cellulose powder (the solid support that binds the target). The 5' end of the isolated mRNA was sequenced by hybridizing it to a CYC1-specific oligonucleotide primer and enzymatically reproducing the isolated sequence in a dideoxy chain termination reaction (i.e., amplification). The multitude of enzymatically produced sequences were detected by gel autoradiography. The dideoxy chain termination reaction clearly constitutes "amplification" in the '338 patent because it produces "additional target, or target-like molecules, or molecules subject to detecting" which are "made enzymatically with DNA...polymerases".

Powell et al. teach the capture of poly$(A)^+$ RNA from a sample by one or two cycles of binding to a probe attached to a solid support, *i.e.* oligo(dT) cellulose (see p. 839, col. 1). After the captured RNA was eluted from the column, it was amplified using the polymerase chain reaction (p. 839, col. 1). Accordingly, this reference teaches each step of claim 1.

Claims 2 and 8, which are dependent on claims 1 and 7, respectively, recite the use of a support that is retrievable. Claim 8 further requires that the amplification product be detected. These claims are anticipated by *Boss et al.*, in which CYC1 mRNA was isolated by hybridization to cloned CYC1 DNA attached to diazobenzyloxymethyl cellulose powder, i.e. a retrievable support and the amplification products were detected by autoradiography. *Powell et al.* also teach the use of a retrievable support, i.e., oligo(dT) cellulose. The amplification product was detected using a dot blot hybridization assay. Accordingly, claims 2 and 8 are anticipated by *Powell et al.*

Each of the *Arsenyan et al.*, *Gaubatz et al.*, *Boss et al.* and *Powell et al.* references disclose the use of a probe on the solid support, *i.e.* an oligo(dT) probe, as claimed in claim 3.

Claim 7, which is directed to a method of detecting a target polynucleotide, recites the same three steps as claim 1, followed by the additional step of detecting the presence of the amplified polynucleotide. *Arsenyan et al.* detected the presence of amplified target polynucleotide by colony hybridization to radioactively labeled 5S RNA. *Gaubatz et al.* detected the amplification product by measuring the amount of tritiated dCTP incorporated during the amplification step. *Boss et al.* detected the amplification product by autoradiography. *Powell et al.* detected the amplified polynucleotide by conducting a dot blot hybridization in which the PCR amplification product was hybridized to radioactively labeled oligonucleotide probes. Therefore Claim 7 is anticipated by each of these references.

Claims 4-6 and 10-12, which depend from claims 1 and 7, respectively, recite that amplification is effected by a polymerase, which can be selected from a group including DNA

polymerase. Reproduction of DNA during the growth of the *E. coli* transformants in *Arsenyan et al.* was perforce carried out by DNA polymerase. *Gaubatz et al.* employed both AMV reverse transcriptase and DNA polymerase (Klenow) in the amplification of target sequence. *Boss et al.* employed reverse transcriptase in the dideoxy chain termination reaction. *Powell et al.* employed *Taq* polymerase to amplify target polynucleotide. Accordingly, the limitations of claims 4-6 and 10-12 are fully met by each of the references.

Claims 13 and 14, which are dependent on claim 7, additionally recite that the amplified polynucleotide is contacted with a label (claim 13), which can be a labeled probe (claim 14). The colony hybridization of the amplified DNA with radioactively labeled 5S RNA in *Arsenyan et al.* meets these limitations. *Powell et al.* used radioactively labeled oligonucleotide probes to detect PCR amplification products.

Claims 24-26 are directed to kits for amplifying a target polynucleotide in a sample comprising means for performing target capture and amplification of the target. In view of the foregoing discussion, it is clear that each of the recited means are disclosed in *Arsenyan et al.*, *Gaubatz et al.*, *Boss et al.* and *Powell et al.*

Claim 27 is directed to a method of amplifying a target polynucleotide in a sample in which the sample is contacted with a capture probe to form a probe-target complex; the sample is then contacted with a solid support that binds the probe-target complex; the support and bound probe-target complex are separated from the sample; the support and bound probe-target complex is contacted with a second medium; the probe-target complex is released into the second medium; the support is removed from the second medium; and the target is amplified.

Arsenyan et al. disclose all of these steps. In *Arsenyan,* a sample containing the (+) strand of 5S DNA target is isolated by contacting it with a poly(A)-tailed 5S RNA probe. The sample containing this probe-target complex is then contacted with a solid support, *i.e.* oligo-dT cellulose, which binds the probe-target complex. The support and bound probe-target complex are separated from the sample by washing the column. The support and bound probe-target complex are contacted with a second medium, *i.e.* elution buffer, into which the probe-target complex is released and the support is removed by allowing the elution buffer to flow out of the column (see p.100, col. 2). After this process is repeated, the isolated DNA is amplified by cloning into *E. coli* to produce a multitude of amplification products in the transformants. Claim 27 is anticipated by *Arsenyan et al.*

Claim 28 is directed to a method of detecting a target polynucleotide in a sample which comprises the steps of claim 27 and the additional step of detecting the presence of the target polynucleotide. Since *Arsenyan et al.* detects the presence of the target sequence by colony hybridization of the *E. coli* transformants (p. 101, col. 1), it also anticipates claim 28.

Claims 29-33, which depend from claims 27 and 28, add the limitation that amplification is effected by a polymerase, which may be a DNA polymerase. Since amplification in *E. coli*, as taught in *Arsenyan et al.* is effected by DNA polymerase, these claims are also anticipated by *Arsenyan et al.*

Claim 34 is directed to a method of amplifying target polynucleotide in which the sample medium is contacted with a solid support and a probe which binds to the target polynucleotide and the support; the support and bound target are separated from the sample medium; the support

and bound probe and target are contacted with a second medium; the target polynucleotide is released into the second medium; the support and bound probe are removed from the second medium; and the target is amplified. *Arsenyan et al.*, *Gaubatz et al.*, and *Powell et al.* each disclose processes in which target polynucleotide is contacted with a solid support and a probe. After the sample was contacted with the solid support and probe, allowing the target polynucleotide to bind thereto, the remainder of the sample was separated from the support and bound probe and target polynucleotide by flowing out of the chromatography column. The column was then contacted with an elution buffer, *i.e.* a second medium, and the target polynucleotide was released into this medium. The elution buffer containing the released target flowed out of the column, thereby separating the support and bound probe from the elution buffer. In each case, the recovered target polynucleotide was then amplified, as described in detail above. In the case of *Boss et al.*, the captured target polynucleotide was separated from the sample by washing the cellulose powder having the support and target sequence bound thereto, eluting with an elution buffer, *i.e.* a second medium, and precipitating with ethanol. Thus, each of these references contains every element of claim 34.

Claims 35-37, which are dependent on claim 34, additionally recite that amplification is effected by the use of a polymerase (claim 35), which can be chosen from a recited group of polymerases (claim 36) that includes DNA polymerase (claim 37). Each of *Arsenyan et al.*, *Boss et al.*, *Gaubatz et al.*, and *Powell et al.* employ a form of DNA polymerase in the amplification step. Accordingly, each of these references anticipates claims 35-37.

Claim 38 is directed to a method for detecting a target polynucleotide, which comprises the steps recited in claim 34 and the additional step of detecting the amplified polynucleotide. As previously indicated, each of *Arsenyan et al.*, *Boss et al.*, *Gaubatz et al.*, and *Powell et al.* discloses the steps recited in claim 34. Moreover, as discussed above in connection with claim 7, each of these references also discloses the detection of amplified polynucleotide. Consequently, each of the references anticipates claim 38.

Claims 39 and 40, which are dependent on claim 38, recite the additional limitation that amplification is effected by a polymerase (claim 39) or a DNA polymerase (claim 40). Since *Arsenyan et al.*, *Boss et al.*, *Gaubatz et al.*, and *Powell et al.* each employ a DNA polymerase for amplification, these claims are anticipated.

Claim 41, which depends from claim 1, recites that amplification is performed *in vitro* to produce a multitude of polynucleotide amplification products. *Gaubatz et al.*, *Boss et al.* and *Powell et al.* each employ *in vitro* amplification to produce a multitude of amplification products. Accordingly, each anticipates claim 41.

Claim 42, which depends from claim 41, recites that amplification is linear or exponential. Claim 43 further limits amplification to exponential amplification. *Powell et al.* discloses PCR amplification of the target polynucleotide, an exponential amplification process (*Mullis*, U.S. Pat. No. 4,683,202). The strand displacement amplification method of *Gaubatz et al.* has an exponential phase (see p. 180, col. 1). Consequently, claims 42 and 43 are anticipated.

Claim 44, which is dependent on claim 41, recites that amplification is carried out using a polymerase and at least one oligonucleotide primer. *Powell et al.* discloses amplification by PCR using two primers and a polymerase. The strand displacement method employed by *Gaubatz et al.* and the amplification method of *Boss et al.* each use one oligonucleotide primer and a polymerase. Therefore Claim 44 is anticipated.

Claim 45 further limits claim 44 by reciting that amplification is linear or exponential. As previously indicated, both *Powell et al.* and *Gaubatz et al.* teach exponential amplification and anticipate the claim.

Claim 46, which depends from claim 41, recites the use of more than one polymerase to amplify the target polynucleotide. Amplification of the target sequence by *Gaubatz et al.* employed more than one polymerase. The first stage of amplification involved producing a cDNA from the captured RNA polynucleotide by using AMV reverse transcriptase, which is an RNA-directed DNA polymerase. In the subsequent strand displacement steps of the amplification process, DNA polymerase is used to reproduce the target polynucleotide. Thus, the claim is anticipated.

Claims 47-51 recite the same limitations recited in claims 41-45, respectively, but depend from claim 7, which is directed to methods of detection. Consequently, they differ from claims 41-45 only in that they incorporate the step of detecting the amplified polynucleotide. Since *Gaubatz et al.* and *Powell et al.* each meet all the limitations of claims 41-45 and also describe detection of the amplified polynucleotide (see *supra*), both references anticipate claims 47-51.

Claim 52, which depends from claim 47, recites amplification using more than one polymerase. As discussed above, *Gaubatz et al.* teach the use of more than one polymerase for amplification and anticipate the claim.

Claims 55, 58 and 59, which depend, respectively, on claims 24, 34 and 38, provide the additional limitation that the means for amplification provide for *in vitro* amplification to produce a multitude of amplification products. *Gaubatz et al.*, *Boss et al.*, and *Powell et al.* each describe *in vitro* amplification that produces a multitude of amplification products. Moreover, each of the references meets all the limitations of claims 24, 34 and 38 (see *supra.*). Consequently, claims 55, 58 and 59 are anticipated by these references.

In summary, based on the foregoing discussion, Protestor submits that:

- Claims 1, 3-7, 10-12, 24-26 and 34-40 are anticipated by each of *Arsenyan et al.*, *Gaubatz et al.*, *Boss et al.* and *Powell et al.*
- Claims 2 and 8 are anticipated by *Boss et al.* and *Powell et al.*
- Claims 13 and 14 are anticipated by *Arsenyan et al.* and *Powell et al.*
- Claims 27-33 are anticipated by *Arsenyan et al.*
- Claims 41, 44, 55, 58 and 59 are anticipated by *Gaubatz et al.*, *Boss et al.*, and *Powell et al.*
- Claims 42, 43, 45 and 47-51 are anticipated by *Gaubatz et al.* and *Powell et al.*
- Claims 46 and 52 are anticipated by *Gaubatz et al.*

IV. ALL NEWLY PRESENTED CLAIMS MUST BE REJECTED BECAUSE THEY ARE NOT SUPPORTED BY THE DISCLOSURES OF THE '338 PATENT.

The reissue application includes new claims 41 to 59, which seek to add various limitations to the amplification step of applicants' method. The new claims depend ultimately from independent claims that were not modified substantively from those of the '338 patent.[10] As discussed above, many of the new claims are directed to embodiments in which the target polynucleotide is "amplified *in vitro* to produce a multitude of polynucleotide amplification products" or in which the amplification is "linear or exponential." Other new claims define "means for" practicing such methods in kits.

Beginning with the filing of the '920 application in December 1987, applicants have consistently defined the term "amplify" very broadly. Applicants' definition literally includes virtually all known ways of producing additional target molecules, target-like molecules, or molecules subject to detection in place of the target molecule (col. 2, ll. 9-19). Applicants' literal definition could be understood to include *in vivo* and *in vitro* amplification, linear and exponential amplification, and target-specific and non-specific amplification that may be

[10] Claims 41-46 depend ultimately from claim 1, claims 47-52 depend ultimately from claim 7, claim 53 depends from claim 19 (which was modified to correctly refer to an antecedent term), claim 54 depends from claim 20, claim 55 depends from claim 24, claim 56 depends from claim 27, claim 57 depends from claim 28, claim 58 depends from claim 34, and claim 59 depends from claim 38.

practiced by using a wide variety of available procedures.[11] Applicants maintained their broad definition of amplification at all times during the prosecution of the '338 patent.

In now seeking to add limitations to the amplification step of the new claims, reissue applicants hope to avoid certain of the prior art references discussed in Sections I-III of this Protest. The prior art references require that the new reissue claims be rejected even if the limitations are considered. Furthermore, the limitations now claimed by reissue applicants are not supported by the disclosures of the '338 patent. The new claims must therefore be rejected on the basis of the "written description" requirement of 35 U.S.C. § 112 and the requirement of 35 U.S.C. § 251 that a reissue application be for the same "invention disclosed in the original patent," without the addition of new matter.

The reissue application defines the term "amplify" very broadly (col. 2, ll. 9-15), so that the proposed limitations would literally apply to all amplification methods. However, the disclosures actually describe only the combination of target capture with *in vitro* amplification wherein the amplification method is non-specific.

[11] Protestor believes that the original claims of the '338 patent, properly construed, do not encompass target-specific amplification in light of the narrow disclosures of the '338 specification, but Protestor reserves more detailed arguments regarding the scope of the original claims for *inter partes* proceedings.

In their detailed description of the invention, the reissue applicants particularly emphasize the benefits of combining target capture with non-specific amplification. For example, the specification states that:

> The sensitivity of the above DNA or RNA target capture methods can be enhanced by amplifying the captured nucleic acids. This can be achieved by *non-specific* replication using standard enzymes (polymerases and/or transcriptases). (Col. 30, ll. 14-18, emphasis added.)

The specification further stresses that non-specific amplification of the purified target nucleic acid employs non-specific enzymes or primers, which can replicate substantially any nucleic acid sequence:

> Amplification of the target nucleic acid sequences, because it follows purification of the target sequences, can employ *non-specific* enzymes or primers... *Thus no specifically tailored primers are needed for each test, and the same standard reagents can be used, regardless of targets.* (Col. 30, ll. 30-40, emphasis added).

Other portions of the detailed description also describe target capture combined only with non-specific amplification. For example, as shown in "FIGS. 4, 5, and 6, the isolated target is non-specifiably [*sic*] amplified" (col. 15, ll. 56-58). FIGS. 3 and 4 depict amplification of target DNA ("substantially free of sample impurities, debri[s] and extraneous polynucleotides") using core RNA polymerase (which transcribes non-specifically). (Col. 15, ll. 59-65.) FIGS. 5 and 6 depict amplification of isolated target DNA using DNA polymerase and "non-specific hexamer primers"(col. 16, ll. 10-29, particularly lines 21-23). The specification also describes "the situation where the target is RNA:... [in which] the target RNA can be replicated nonspecifically by denaturing the RNA and subjecting the RNA to an enzyme such as Qβ replicase or reverse transcriptase" (col. 16, ll. 5-9).

All of the Examples that include amplification describe *only* non-specific amplification, using methods that were known in the prior art. That is, all of the described embodiments[12] use non-specific amplification of a target polynucleotide that has been captured from the sample. Therefore, taken as a whole, the specification teaches only the combination of target capture and *in vitro* non-specific amplification, despite the Reissue Applicants' broad literal definition of "amplify" at column 2, lines 9-19.

Reissue applicants now seek to add certain limitations to the amplification step of their claims. If applicants' claims are to be narrowed, they must be narrowed in a manner consistent with applicants' actual disclosures in the specification. Therefore, the new claims must be to amplification methods actually disclosed in the specification. For example, any limitation to *in vitro* amplification must be combined with a limitation to non-specific amplification because *in vitro* amplification is not mentioned anywhere in the specification except in connection with non-specific amplification.

35 U.S.C. § 112 requires the specification to contain a written description of the invention that is a full, clear, and concise description of the invention. The written description must be equivalent to the claimed subject matter. *Lockwood v. American Airlines, Inc.*, 107 F.3d 1565, 1571-1572, 41 USPQ2d 1961, 1966 (Fed. Cir. 1997). The disclosure must permit one skilled in the art to reasonably conclude that the inventor had possession of the invention *as claimed. Vas-Cath, Inc. v. Mahurkar*, 935 F.2d 1555, 1563-4, 19 USPQ2d 111, 1117 (Fed Cir. 1991). With respect to claim limitations, the written description must clearly convey to one

[12] Where the examples of the specification reflect the only teaching of the specification, those examples are not merely "preferred embodiments," but constitute the applicant's only disclosed invention. *Wang Laboratories Inc. v. America Online Inc.*, 197 F.3d 1377, 1384, 53 USPQ2d 1161, 1165 (Fed. Cir. 1999); *General American Transportation Corp. v. Cryo-Trans, Inc.*, 93 F.3d 766, 770, 772, 39 USPQ2d 1801, 1803, 1805-06 (Fed. Cir. 1996).

skilled in the art that the applicant made the invention having those limitations. *Martin v. Mayer*, 823 F.2d 500, 505, 3 USPQ2d 1333, 1337 (Fed. Cir. 1987).

Separately from Section 112, 35 U.S.C. § 251 requires that reissue claims be for the same "invention disclosed in the original patent," without the introduction of new matter. Pursuant to sections 112 and 251, limitations added in the reissue application must be rejected if not supported by, and consistent with, the applicants' actual disclosures as set forth in the '338 patent.

The specification contains only two possible sources for disclosures that can serve to limit applicants' amplification step. Neither source supports the limitations proposed by the new claims of the reissue application.

The first possible source is applicants' definition of the term "amplify." However, applicants have so broadly defined that term that it does not disclose the limitations of reissue applicants' new claims. Applicants are precluded from now modifying their long-standing definition in order to narrow its meaning. While an applicant may choose to define a term for the purposes of its use in an application, the applicant must use the term *consistently* in the specification and claims and any special meaning must also be *consistently* adhered to in determining patentability and validity. *Burlington Industries, Inc. v. Dayco Corp.* , 849 F.2d 1418, 1421, 7 USPQ2d 1158 (Fed. Cir. 1988); 5A *Chisum On Patents* § 18.03[3][c], pp. 18-159 to 18-160 (1999). In this regard, an inventor's right to define the terms used in the application ends when the patent issues and the application acquires its own independent life as a technical disclosure to the public. *Lear Siegler, Inc. v. Aeroquip Corp.*, 733 F.2d 881, 888-889, 221 USPQ 1025, 1031 (Fed. Cir. 1984).

The second possible source for the new claim limitations is the detailed description of the invention including the Examples contained in the specification. Apparently recognizing that they may not simply change their definition of "amplify," reissue applicants in fact rely on the Examples to support their additional proposed limitations. See Preliminary Amendment and Remarks at p. 10.

While the examples disclose *in vitro* amplification, each of the examples makes this disclosure only in connection with non-specific amplification. In each instance, the *in vitro* amplification method described in the Examples is non-specific amplification. Therefore the only disclosure of *in vitro* amplification is a disclosure of *in vitro* non-specific amplification.

Reading the actual disclosures of the specification, one skilled in the art would discern that applicants' invention was limited to *in vitro* amplification using only methods of non-specific amplification. Although reissue applicants may contend that other forms of amplification (e.g., target-specific amplification) are obvious, no other amplification methods are disclosed in the specification. The adequacy of applicants' disclosure must focus on what the specification *actually discloses*, not what applicants contend might be obvious from the specification. *Lockwood v. American Airlines, Inc.*, 107 F.3d 1565, 1571-1572, 41 USPQ2d 1961, 1966 (Fed. Cir. 1997).

There are numerous methods of *in vitro* amplification and many of these produce "a multitude of amplification products." To the extent the Examples suggest limitations, they suggest multiple, inseparable limitations. The specification only discloses *in vitro* non-specific amplification. Claims that do not concurrently contain both of these limitations are inherently overbroad. Applicants are not free to now modify their broad definition of "amplify" directly, nor are they now free to indirectly modify that definition by selectively claiming some, but not all, of the limitations suggested by the Examples.

To comply with the requirements of 35 U.S.C. § 112 and § 251, any limitation of the new claims based on the disclosures of the Examples must include a limitation to non-specific amplification. The new claims, as presently stated, are not supported by the actual disclosures of the '338 patent and must be rejected under the "written description" requirement of 35 U.S.C. § 112, first paragraph and the "same invention" requirement of 35 U.S.C. § 251.

CONCLUSION

Protestor respectfully requests that the Examiner consider the above remarks when examining the reissue application of the '338 patent. Considered together, the above remarks demonstrate that:

- All claims are invalid as obvious under 35 U.S.C. § 103;
- Many claims are invalid as anticipated under 35 U.S.C. § 102; and
- The claims added to the reissue application should be rejected under the "written description" requirement of 35 U.S.C. § 112, first paragraph and the requirement of 35 U.S.C. § 251 that the reissue application be for the "invention disclosed in the original patent."

Respectfully submitted,

Date: August 1, 2000

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IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

In re Reissue Application of: U.S. Patent No. 5,750,338 Mark L. Collins *et al.* Reissue Serial No. 09/533,906 Reissue Application Filed: March 8, 2000 For: TARGET AND BACKGROUND CAPTURE METHODS WITH AMPLIFICATION FOR AFFINITY ASSAYS	Group Art Unit: Unassigned Examiner: Unassigned

DECLARATION OF MICHAEL M. HARPOLD, Ph.D.

ATTENTION: REISSUE LITIGATION BOX 7
Assistant Commissioner for Patents
Washington, D.C. 20231

Sir:

I, Michael M. Harpold, residing at 5920 N. Placita Tecolote, Tucson, Arizona 85718, do hereby declare as follows:

1. I have been Principal of EnkephaSys, involved in biomedical consulting, since 1999. From 1998-1999, I was Chief Scientific Officer and a Director of the National Center for Genome Resources, a non-profit research institute involved in computational biology and bioinformatics research. I was employed by SIBIA Neurosciences, Inc. (formerly The Salk Institute Biotechnology/Industrial Associates, Inc.) from 1981, serving as a founding Senior Staff Scientist from 1981-1982, as Director of Scientific Planning from 1982-1986, as Vice President for Scientific Planning from 1986-1990, and as Vice President, Research from 1990-1998. From

1973-present, I have conducted and directed research in a variety of areas of biological science, including molecular biology, nucleic acid hybridization, and DNA probe technologies. A copy of my *curriculum vitae* is attached hereto as Exhibit A.

2. I have read copies of the following documents relating to U. S. Patent No. 5,750,338 (the '338 patent'), which is the subject of the above-captioned reissue application:

U.S. Patent No. 5,750,338;

Prosecution file history of U.S. Serial No. 08/238,080, including the Declaration of David H. Persing, M.D., Ph.D., filed July 14, 1997; and

Preliminary Amendment filed with reissue application no. 09/533,906, dated March 8, 2000.

3. The '338 patent describes methods of detecting nucleic acid sequences that use capture of polynucleotide sequences on a solid phase support and non-specific amplification of the captured polynucleotide. I have been informed that the filing date of the first patent application that discloses this combination of steps and from which the '338 patent claims priority is December 21, 1987 (the "filing date").

4. I believe that the invention claimed in the '338 patent and its reissue application was known or obvious to one skilled in the art at the filing date of the application. I also believe that the specification of the '338 patent fails to convey to one of ordinary skill in the art that the inventors were in possession of the invention claimed in new claims 41-59. More specifically, the specification conveys that the applicants were only in possession of a method that employed *in vitro* non-specific amplification, whereas new claims 41-59 are not limited to non-specific amplification. In support of these opinions, in the following paragraphs, I will present a short

summary of the general state of the art at the filing date of the '338 patent application; my view of the level of skill in the art at the filing date of the '338 patent application; a discussion of disclosures in published references that were available to those skilled in the art at the filing date of the '338 patent application; and a discussion of what the specification would convey to one of skill in the art regarding the scope of the methods that the reissue applicants were in possession of at the filing date. I will also present my views of the state of the art in contrast to those expressed by Dr. David Persing in his declaration filed in 1997.

5. General State of the art at the filing date of the '338 patent application.

Following the discovery of reverse transcriptase (1970), and the development of cloning (1973) and DNA sequencing (1975), biological scientists began to study individual genes and DNA sequences associated with gene expression. It also became possible to develop DNA probe based methods for the detection of normal and mutant genes and for the detection and identification of infectious organisms using the techniques of molecular biology. The low frequency of individual genes in the genome (e.g., 1 in 40,000 human genes) and/or the low concentration of individual gene expression products in soluble cellular components presented technical problems for such studies and methods. Therefore, scientists were motivated to develop methods to separate and/or concentrate individual genes from other cellular components. In addition, scientists were motivated to amplify individual sequences to provide sufficient quantities for analysis and detection. Recombinant DNA cloning was one method that allowed scientists to isolate discrete DNA fragments from any organism and produce (amplify) large amounts of the same DNA for further characterization. Cloning itself was, however, a very laborious process when genomic DNA, or DNA transcribed from unfractionated messenger RNA, were used as the starting material. Often, tens of thousands or hundreds of thousands of

clones had to be screened to find those containing the desired sequence. See, *e.g.*, Maniatis et al., Molecular Cloning A Laboratory Manual (Cold Spring Harbor Laboratory, Cold Spring Harbor, NY 1982), in Chapter 10, pages 309-362.

To increase the frequency of clones with a particular target sequence, scientists often isolated mRNA from tissue that specifically produced the target gene product. For example, mRNA was isolated from pancreatic islet cells to enrich for sequences encoding insulin. Methods were developed to separate mRNA from other RNA and proteins found in cellular extracts, such as by using hybridization of the poly(A) tail of mRNA to a complementary oligo(dT) sequence in chromatography, *i.e.*, capture on a solid support (see Maniatis et al., *supra*, Chapter 6). The isolated mRNA could then be converted to cDNA using reverse transcriptase, cloned and amplified by replication in bacterial cells. Thus, it was commonplace before the filing date for scientists to employ the combination of enrichment by target capture and amplification in order to generate detectable amounts of nucleic acid sequences of interest.

The application of molecular biological techniques to the diagnosis of disease has been recognized for decades. Many commonly used immunoassays for infectious agents employ a combination of target capture and amplification to enhance detection sensitivity. In so-called sandwich assays, a solid support containing a bound capture antibody is used to capture an antigen from the specimen being tested. The other sample components are washed away. Other antibodies, coupled with enzymes, bind to the captured antigen. After excess detection antibodies are washed away, an amplified signal is generated from the bound enzyme by its repeated production of a readily detected product. Immunoassays are members of the general class of ligand binding assays, in which a component characteristic of the infectious agent is bound specifically to a support (target capture) and detected. In cases where direct detection is

insufficiently sensitive, various amplification methods are commonly employed to amplify the signal generated from the presence of the captured ligand. Those skilled in the art recognized that hybridization assays were another form of ligand binding assay and that unique nucleic acid sequences present in infectious agents could be used to detect and identify them in the same way in which antigen:antibody reactions were employed. In these assays, which were commonly performed in a sandwich format, a labeled nucleic acid probe was used to detect the presence of a nucleic acid sequence indicative of the presence of the infectious organism.

In some infectious diseases, the number of organisms present may be small, and test sensitivity must be high to detect the majority of cases. As with immunoassays, scientists found that diagnostic tests based on hybridization between complementary DNA sequences often lacked sensitivity when the number of infectious agents in the sample was small because the signal over background that could be generated from a low number of hybrids was insufficient for reliable detection. Thus, hybridization assays were insensitive compared to tests in which the number of organisms was amplified by growing them in culture. To overcome the lack of sensitivity of DNA-based probe tests, scientists used amplification to enhance the signal obtainable from the probe:target hybrids. As with the antibodies used in immunoassays, probes could be labeled with enzymes. The enzyme label would repeatedly transform a substrate into product, thereby amplifying the signal from each hybrid formed. In this way, the sensitivity of detection was enhanced.

Thus, it was common practice in the art prior to December, 1987, to combine technologies, such as target capture, amplification and hybridization methods, to obtain specific and sensitive detection of sequences of interest.

6. I consider the level of ordinary skill in the art of molecular biology at the filing date of the '338 patent application to have been that of an individual with a Ph.D. in the biological sciences and two years of postdoctoral experience. Such experience would have allowed the individual to develop the skills of a molecular biologist or genetic engineer using the techniques of DNA and RNA isolation and characterization, cDNA synthesis, cloning, liquid and solid phase hybridization (including knowledge of the conditions influencing hybrid formation and stability), affinity chromatography, isotopic and non-isotopic labeling methods, DNA sequencing methods, and nucleic acid amplification, including, but not limited to, PCR.

7. Relevant techniques that were well known in the art at the filing date of the '338 application, based on published scientific articles and U.S. Patents, are summarized below. They have been grouped into sections related to Capture Technology, Amplification Technology, and the combination of Capture and Amplification Technology. These provide a more detailed basis for my opinion that one skilled in the art at the filing date of the '338 application would have been motivated to combine known techniques to produce the invention claimed in the '338 patent.

8. Capture Technology

Before the filing date, it was well known among those skilled in the art that a polynucleotide of interest could be purified from a sample by contacting it with a solid support that would bind the polynucleotide and then separating the bound target from the sample. Such a purification step would remove components in the sample that would otherwise prevent or inhibit the further manipulation (e.g., cloning or in vitro amplification) of the target polynucleotide.

It was standard practice for many years before 1987 to isolate poly(A)-tailed mRNA from a biological sample by contacting the sample with oligo(d)T bound to a solid support (e.g., cellulose) under conditions that permitted hybridization of the poly(A)$^+$ mRNA with the oligo(dT) moiety (see Maniatis et al., *supra*, Chapter 6). After separating the bound support and mRNA from the sample, the purified mRNA could then be released from the solid support and used in a variety of ways (e.g., in cDNA synthesis and/or molecular cloning). For example, Gaubatz et al., *Biochim. Biophys. Acta* 825:175-187 (1985) described the isolation of poly(A)$^+$ mRNA using oligo(dT)-cellulose chromatography followed by conversion of the purified mRNA into cDNA and further amplification using the Klenow fragment of DNA polymerase I. It was also well known in the art before December 1987 that sequence-specific capture of a target polynucleotide could be carried out by contacting a sample containing the target polynucleotide with a solid support and a capture probe that was capable of binding the solid support and that contained a sequence of nucleotides complementary to a specific sequence in the target polynucleotide. For example, *U.S. Patent No. 4,672,040* (Josephson) and *U.S. Patent No. 4,554,088* (Whitehead et al.) describe use of dispersible solid supports--magnetic beads--with bound polynucleotide capture probes for isolating target polynucleotides. By separating the magnetic beads with their adherent target polynucleotide using a magnet, the target polynucleotide is separated from other sample components capable of inhibiting or negatively affecting further manipulation, including detection, of the target polynucleotide.

Nucleic acid hybridization assays employing sequence-specific capture of target polynucleotides were also well known in the art before December 21, 1987. In these assays, target polynucleotide was first isolated from a sample by contacting the sample with a solid support having bound to it a capture probe with a sequence complementary to at least a portion

of the target under conditions that permit the target and probe to hybridize. After separating the bound target from the sample, the target could be detected (e.g., by using a labeled probe). Such assays are described in Polsky-Cynkin et al., *Clin. Chem.* 31(9):1438-1443 (1985) and *U.S. Patent No. 4,563,419* (Ranki et al.). Syvänen et al., *Nucleic Acids Res.*, 14: 5037-5048 (1986), describe a sandwich hybridization assay in which a biotin-containing capture DNA and ^{125}I-labeled probe were first hybridized to the target polynucleotide in solution and the target-probe-capture DNA complex was subsequently contacted with a solid support (i.e., streptavidin-agarose) and bound to the support via a streptavidin-biotin linkage through the biotinylated capture probe. The presence of the ^{125}I-labeled probe allowed detection of the bound complex. Because hybridization of probe to the target occurred in solution prior to capture on the solid support, greater capture efficiency was achieved using solution-phase hybridization kinetics.

9. Amplification Technology

Various methods for amplifying nucleic acids were well known in the art prior to the filing date. One skilled in the art would understand "amplify" as defined in the '338 patent (col. 2, lines 9-19) to encompass a wide variety of well known methods to either directly produce more copies of the captured polynucleotide or indirectly amplify the presence of the target polynucleotide. Direct amplification methods would include known methods of *in vitro* nucleic acid amplification, including cDNA synthesis, and cloning (i.e., additional target molecules are produced by inserting the target nucleic acid into host cells and growing the transformed cells). An example of indirect amplification would be cell-free gene expression (i.e., protein molecules are created by virtue of the presence of the target polynucleotide). In addition to these molecular biology techniques, other methods of target amplification were known before December 1987. In particular, all of the methods of amplification disclosed in the '338 patent were well known

then. Feinberg et al., *Anal. Biochem.* 132:6-13 (1983) had described use of random hexamer primers to initiate non-specific enzymatic amplification of polynucleotides (Examples 5 and 6 of the '338 patent). Blumenthal et al., *Proc. Natl. Acad. Sci. U.S.A.* 77:2601 (1980) had described use of the enzyme Q-beta (Qβ) replicase to amplify polynucleotides (Example 7 of the '338 patent). R. Burgess in RNA Polymerase (Cold Spring Harbor Press, (1976), pp. 69-100 had described use of RNA polymerase core enzyme to non-specifically replicate polynucleotides (Example 4 of the '338 patent). Besides the amplification methods described in the '338 patent, other methods of amplifying polynucleotides were well known in the art before the filing date. Gaubatz et al., *Biochim. Biophys. Acta* 825:175-187 (1985) had described a method of amplifying cDNA sequences by a polymerase-mediated strand displacement synthesis. PCR had been described in a number of publications before the filing date and in U.S. patents filed before December, 1987, including U.S. Patent No. 4,683,202 (Mullis).

10. Combination of Capture and Amplification Technology

In my opinion, the scientific literature had also taught prior to the filing date that it would be useful to combine the techniques of target capture and amplification to enrich and detect target molecules of interest in samples, including the capture and amplification of polynucleotides.

As discussed above, solid supports were widely used to isolate nucleic acids to study their structure and function or to detect the presence of a particular nucleic acid in a sample. Because the amount of a target polynucleotide in a sample is often very small, the desirability of increasing the proportional amount of isolated polynucleotide before further analysis or detection would have been obvious to one skilled in the art. It was common practice in the art to capture small amounts of mRNA on solid supports, convert the mRNA to cDNA using reverse

transcriptase, an RNA-dependent DNA polymerase, and then further amplify the cDNA by cloning prior to detecting and/or analyzing the DNA. Once methods of *in vitro* amplification became widely available, it was obvious to combine these techniques with target capture on solid supports. Moreover, because target capture was known to be less than 100% efficient, it would have been obvious to one skilled in the art to combine it with amplification procedures to increase the amount of captured target (i.e., to compensate for any losses during the capture step) before detection.

Brown et al., in "Methods of Gene Isolation" *(Ann. Rev. Biochem.,* 43:667-693 (1974)), clearly recognized the desirability of combining target capture on solid supports with amplification. The authors reviewed a number of methods for isolating nucleic acids of interest, and devoted one section to the use of polynucleotides fixed to insoluble matrices to isolate DNA. At pages 673-674, Brown et al. describe using RNA or DNA fixed to an insoluble support to effect sequence-specific isolation of target DNA containing a sequence complementary to the probe. In their "Concluding Remarks" (p. 687) Brown et al. state that purification of DNA (*e.g.* using polynucleotides fixed to insoluble matrices) could be coupled with a method by which, "...a small amount of a given gene can be increased enormously in amount" such as by an "amplification step ... carried out *in vitro* by an efficient DNA polymerase, which would replicate faithfully each molecule of DNA many times."

Arsenyan et al., *Gene* 11:97-108 (1980) also recognized the desirability of combining target capture with amplification because they described a method to produce "amplified homogeneous DNA sequences" for the purpose of studying gene arrangement. Their process captured complementary single strands of the 5S RNA gene by hybridization to probes affixed to cellulose supports followed elution of the captured strands, annealing of the strands to produce

double stranded DNA which was inserted into a cloning vector, transformation of host cells with the vector and amplification of the DNA by growing the transformed host cells. At the time of filing of the '338 patent, it would have been obvious to one of ordinary skill in the art that other known amplification methods could have been substituted for the cloning step of *Arsenyan et al.* The obviousness of such substitution is suggested by the reissue applicants' own definition of "amplify" in the Background of the Invention section of the '338 patent (col. 2, ll. 9-19). In my opinion, substituting other methods of amplification, as they became available, for amplification by cloning would have been obvious and routine to one skilled in the art.

Gaubatz et al., *Biochim. Biophys. Acta* 825:175-187 (1985) described the isolation of poly(A)$^+$ mRNA by using oligo(dT)-cellulose chromatography followed by conversion of the purified mRNA into cDNA and amplification of the cDNA using a polymerase-mediated strand displacement procedure.

Powell et al., *Cell* 50:831-840 (1987) described isolating poly(A)$^+$ RNA by one or two cycles of oligo(dT)-cellulose chromatography to capture Apo-B mRNAs followed by synthesis of cDNA and PCR amplification of the cDNA and detection of specific Apo-B48 sequences.

11. Summary of my opinion based on the state of the art at the time the '338 application was filed.

It is my opinion that the technology described in the '338 patent would have been obvious to one of ordinary skill in the art. Specifically, before the filing date of the '338 patent, it was common practice to isolate a target polynucleotide from a sample by contacting the sample with a solid support which bound to the target polynucleotide either directly or indirectly. The support with its attached polynucleotide and bound target polynucleotide was separated from

the sample, effectively separating the target from other sample components that could interfere with further manipulation of the target polynucleotide. It was also well known in the art that amplifying the target polynucleotide directly or by producing an amplified signal based upon the presence of the target polynucleotide could increase detection sensitivity. It is my opinion that it would have been obvious to one of ordinary skill in the art in December 1987 to perform the combination of those manipulations for the purpose of detecting the presence of a target polynucleotide in a sample. Based upon my understanding of the skill of the art at the filing date of the '338 patent, I conclude that one of ordinary skill would have been motivated to combine both target polynucleotide capture and amplification to ensure success in a detection assay.

12. Analysis of Dr. Persing's Declaration

I have been informed that allowance of the '338 patent followed shortly after the filing of Dr. Persing's declaration. I respectfully submit that Dr. Persing's declaration does not fully characterize the state of the art at the filing date of the '338 patent application. Further, the implicit description of the individual of ordinary skill in the art is inaccurate because it is too low. As discussed above, the skilled practioner at that time would have been aware of the different methods for target capture and amplification and would have been motivated to combine these techniques.

In my opinion, in addition to incorrectly characterizing the state of the art and of the skill level of practioners, Dr. Persing also incorrectly characterized what was known about PCR at the filing date of the '338 application. In paragraph 12 of the declaration, Dr. Persing asserts that, at the filing date, "...[T]hose who were adding amplification to their hybridization assays had a strong incentive to avoid the addition of target isolation steps to their hybridization assays". He bases this statement on his belief that users of PCR believed that PCR was so highly specific that

additional steps for isolating the target were not needed. Dr Persing stated that "It was not until much later that it became apparent that non-specific amplification was occurring despite the careful selection of primers." Dr. Persing was incorrect in stating that the problem of non-specific amplification in PCR had not been recognized by December, 1987, because the PCR inventor himself discussed the problem of "background" amplification[1]. Even if Dr. Persing's statement is accepted as correct, it does not, in my opinion, justify a conclusion that those skilled in the art had no incentive to combine target capture and amplification as described and claimed in the '338 patent.

I have carefully reviewed the specification and claims of the '338 patent and found that PCR amplification is never mentioned or described. Likewise, there is no mention of problems with PCR and no teaching, nor showing, that a combination of target capture and PCR amplification provides any improvement over the PCR process alone. Unlike PCR, all of the amplification methods disclosed in the '338 patent are non-specific amplification methods which would be expected to amplify indiscriminately any polynucleotides present in a sample. Contrary to Dr. Persing's conclusion, one skilled in the art would have had a strong incentive to isolate the target sequence before amplification, as described in the '338 patent, to avoid non-specific amplification of non-target sequences. Thus, for example, one would be motivated to

[1] In U.S. Patent No. 4,683,202, which is directed to the PCR process, inventor Mullis recognized that non-specific amplification of non-target sequences (background amplification) could be a problem, particularly when one is attempting to amplify a single-copy gene. In Example 10, he describes overcoming this problem through the use of nested primers. He also states (col. 5, ll. 34-37) that the starting nucleic acid may be in a purified form. His recognition of the problem directly contradicts Dr. Persing's statement in paragraph 12 of the declaration. Mullis described one approach to overcoming the problem. Having recognized the problem, it is my opinion that other approaches would have been obvious to the average skilled worker, including purification of the target prior to amplification using art-known techniques such as target capture on a solid support.

combine the art-known methods of isolating target polynucleotide on solid supports with the known amplification methods disclosed in the '338 patent.

In paragraph 13 of Dr. Persing's declaration, he states that practitioners of hybridization assays were reluctant to use hybridization techniques to purify their intended targets prior to amplification because the lack of complete binding efficiency of probe to target would result in a reduction of available target, which might be at a low initial concentration in the sample. In my opinion, inefficient binding of capture probe to target does not justify a conclusion that those skilled in the art would be led away from combining target capture and amplification. Although capture methods were not expected to be 100% efficient and the total number of target polynucleotides in an enriched sample would be somewhat reduced (i.e., some would not be captured), the published levels of amplification far exceeded the amount lost during target capture. For example, one might expect to capture only 50 % of target molecules, but amplification typically resulted in a 100-fold increase in the amplified sequences. Therefore, target capture of 50% and 100-fold amplification would result in a 50-fold overall increase in the number of target polynucleotides compared to those in the original sample.

Because of the anticipated loss of target during the capture step, one skilled in the art would have been *motivated* to amplify the captured target prior to detection, thereby increasing the sensitivity of the assay. Brown et al., *supra*, Gaubatz et al., *supra*, and Powell et al., *supra*, all clearly recognized that target capture combined with amplification would be advantageous for providing adequate amounts of the target polynucleotide for analysis.

In paragraph 14 of Dr. Persing's declaration, he states that the claimed invention provided an additional advantage that was unexpected before the filing date, *i.e.* the elimination

of amplification inhibitors present in the sample. In my opinion, one of ordinary skill in the art would have been fully aware before the filing date that isolation of target polynucleotide on a solid support would have advantages, including changing buffer (medium) components and removing substances that would inhibit amplification of the target polynucleotide. The presence of inhibitors of restriction endonucleases or of DNA or RNA polymerases (used in amplification reactions) in biological, environmental, or clinical samples was well known in the art before 1987. Consequently, before then it was standard practice in molecular biology to purify nucleic acids before restriction endonuclease cleavage or polymerase-mediated amplification. It would have been obvious to one skilled in the art that nucleic acid purification, *e.g.* using the known technique of target capture on a solid support, could be desirable before any amplification step which employs a polymerase because it would remove potential inhibitors of the amplification reaction. Exemplary references that discuss enzymatic inhibitors and the need for nucleic acid purification include DNA Synthesis, Kornberg, A., (Freeman & Co., San Francisco 1974) p. 65; Maniatis et al., *supra,* Chapters 6 and 7, pp. 182-196 and 213.

13. Summary of my opinion regarding Dr. Persing's Statements and Conclusions

Based on my knowledge of the state of the art at the time the '338 application was filed, the level of skill of an ordinary practitioner of molecular biology at that time, and the references discussed herein, I believe that Dr. Persing incorrectly characterized the state of the art relevant to the claimed invention of the '338 patent and further incorrectly characterized known aspects of PCR amplification.

14. The '338 patent specification describes methods that are different than those claimed in new claims 41-59.

I have been informed that the patent statutes require that the invention claimed in a patent be described in the patent specification in a manner that reasonably conveys to one of ordinary skill in the art that the inventor was in possession of the invention, as claimed, at the time of filing of the application. I have carefully read the specification and claims of the '338 patent, and it is my opinion that one skilled in the art would conclude from the specification that the inventors were not in possession of the invention of claims 41-59. At most, the specification conveys that the inventors were in possession of a method in which target capture was combined with an *in vitro non-specific* amplification step.

Claims 41-59 are directed to methods in which the amplification step is conducted *in vitro*. None of these claims is further limited to amplification that is non-specific. In the Preliminary Amendment filed with the reissue application, applicants referred to the Examples as support for these new claims. However, the *only* methods of amplification described in the Examples (as well as in the Summary of the Invention and Detailed Description, col.15, l. 56-col. 16, l. 29) are non-specific amplification methods, *i.e.* methods that indiscriminately amplify any nucleic acids present in a sample. Based on the Summary of the Invention, Detailed Description and the Examples, I believe that one skilled in the art would have thought that the only invention that the applicants possessed was one that combined target capture with non-specific amplification methods.

This is further supported by the description at column 30, lines 30-40 of the '338 patent in which the applicants indicate that a benefit of their invention is that it allows the use of non-specific amplification, i.e., the same enzymes and primers can be used for all target sequences. Those skilled in the art at the filing date of the '338 patent would have appreciated that this would have made the assay simpler and less expensive because individual primers would not have to be designed, synthesized and tested for each target (as, for example, in PCR). Specifically, the applicants stated:

> Amplification of the target nucleic acid sequences, because it follows purification of the target sequences, can employ non-specific enzymes or primers... Thus no specifically tailored primers are needed for each test, and the same standard reagents can be used, regardless of targets.

The examples in the specification that describe methods combining target capture with amplification (i.e., Examples 4 to 7) describe several different methods of amplification. The common feature of all of those methods is the non-specific nature of the amplification, a point that is repeatedly emphasized in the specification.

It is also important to note that, although PCR, a primer/target-specific amplification was well known by December 1987, the applicants did not include it in their examples or reference PCR elsewhere in the specification. This is consistent with the conclusion that the inventors only possessed methods that combined target capture with non-specific amplification. Further, no other sequence-specific amplification method is included in the description of the invention and the specification teaches the benefit of using amplification methods that do not require specific primers.

In summary, it is my opinion that one of ordinary skill in the art would conclude from the specification of the '338 patent that the only method the inventors possessed that combined *in vitro* amplification with target capture was a method that used non-specific amplification. Accordingly, claims 41-59 must include a limitation to non-specific amplification or they will be inconsistent with the applicant's disclosed invention, as one skilled in the art would understand it from the specification.

15. Conclusion. It is my opinion that the invention claimed in the '338 patent and its reissue application was known or obvious to one of ordinary skill in the art of molecular biology at the time of the '338 patent application, and that this patent should not be reissued. Further, I believe that the statements and conclusions made in Dr. Persing's declaration of 1997 should not be relied upon as an accurate characterization of the state of the art at the filing date of the '338 patent application. Finally, it is my opinion the 09/533,906 reissue application fails to describe the invention of claims 41-59 because those claims are not limited to the use of non-specific amplification, as described in the specification.

I hereby declare that all statements made herein of my own knowledge are true and that all statements made on information and belief are believed to be true; and further that these statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment or both, under Section 1001 of Title 18 of the United States Code.

July 31, 2000
Date

Michael M. Harpold

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

In re Reissue Application of: U.S. Patent No. 5,570,338 Mark L. Collins *et al.*	)	Group Art Unit: 1634
Reissue Serial No. 09/533,906	)	Examiner: *[Unassigned]*
Reissue Application Filed: March 8, 2000	)	
For: TARGET AND BACKGROUND CAPTURE METHODS WITH AMPLIFICATION FOR AFFINITY ASSAYS	)	Atty. Docket No. Collins Reissue Protest

PROTEST UNDER 37 C.F.R. § 1.291(a)

EXHIBIT A

Curriculum Vitae of Michael M. Harpold, Ph.D.

Curriculum Vitae

MICHAEL M. HARPOLD, Ph.D.

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Chief Scientific Officer and Director, National Center for Genome Resources, Santa Fe, NM (1998-1999)

Vice President, Research, SIBIA Neurosciences (formerly known as The Salk Institute Biotechnology/Industrial Associates, Inc.), La Jolla, CA (1990-1998)

Vice President, Scientific Planning, The Salk Institute Biotechnology/Industrial Associates, Inc., La Jolla, CA (1986-1990)

Director of Scientific Planning and Molecular Biology, The Salk Institute Biotechnology/Industrial Associates, Inc., La Jolla, CA (1982-86)

Senior Staff Scientist, The Salk Institute Biotechnology/Industrial Associates, Inc., La Jolla, CA (1981-82)

Senior Scientist, La Jolla Biological Laboratories, The Salk Institute, La Jolla, CA (1981-1983)

Member, Norris/USC Comprehensive Cancer Center, Los Angeles, CA (1979-1981)

Assistant Professor, Department of Biochemistry, University of Southern California School of Medicine, Los Angeles, CA (1979-1981)

Awards/Honors:

NIH Graduate Fellowship, The Marine Biological Laboratory, Embryology Course, Woods Hole, MA (1974)

Helen Hay Whitney Fellowship, The Rockefeller University (1976-1979)

University Teaching:

TCU, 1971
Teaching Assistant, General Biology Laboratory

Tulane University, 1972-1974
Teaching Assistant, Genetics
Teaching Assistant, Developmental Biology Laboratory
Lecturer, Chemical Embryology

University of Southern California School of Medicine, 1979-1981
Proctor, Medical Biochemistry, Year I Discussion Group
Lecturer, Genetic Engineering-Medical Biochemistry
Lecturer, Graduate General Biochemistry
Course organizer, Lecturer, Molecular Mechanisms of Gene Expression and Cellular Differentiation

Research Support:

Helen Hay Whitney Foundation, Postdoctoral Fellowship, 1976-1979

American Cancer Society, Institutional Research Grant, 1980-1981

National Institute of Child Health and Human Development, NIH, RO-1 Research Grant, 1980-1983

Research support for multiple research projects from various corporations, including Phillips Petroleum, Diamond Shamrock, Hoffmann La Roche, Eli Lilly, and Novartis/Ciba-Geigy, 1982-1998

Scientific Review:

Grants: NIH Biomedical Sciences Study Section, 1982-1983

Journals: Proceedings National Academy of Sciences, USA, J. Neurochemistry, J. Biological Chemistry, Analytical Biochemistry, Neuropharmacology, J. Pharmacology and Experimental Therapeutics, J. Molecular Neuroscience, Cell, Genomics, Nucleic Acids Research

Publications:

M.M. Harpold and S.P. Craig. (1975) A possible mechanism for regulation of mitochondrial RNA polymerase during development of the sea urchin embryo. *J. Cell Biol.* 67, 157.

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Patents:

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S.B. Ellis and M.M. Harpold. (1988) Nucleic acid probes for prenatal sexing. *US Patent* 4769319.

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M.A. Akong, M.M. Harpold, G. Velicelebi, and P. Brust. (1997) Automated analysis equipment and assay method for detecting cell surface protein and/or cytoplasmic receptor function using same. *US Patent* 5670113.

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IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

In re Reissue Application of: U.S. Patent No. 5,750,338 Mark L. Collins *et al.* Reissue Serial No. 09/533,906 Reissue Application Filed: March 8, 2000 For: TARGET AND BACKGROUND CAPTURE METHODS WITH AMPLIFICATION FOR AFFINITY ASSAYS	Group Art Unit: 1634 Examiner: *[unassigned]*

INFORMATION DISCLOSURE STATEMENT ACCOMPANYING PROTEST UNDER 37 CFR § 1.291(a)

ATTENTION: REISSUE LITIGATION BOX 7
Assistant Commissioner for Patents
Washington, D.C. 20231

Sir:

The following information is brought to the attention of the Examiner. The items listed on the attached form PTO-1449, submitted pursuant to 37 C.F.R. 1.291(b)(1), are relied on in the protest filed herewith. Copies are enclosed for the convenience of the Examiner.

The following is a concise explanation of the relevance of each of the listed items.

CERTIFICATE OF DELIVERY

I hereby certify that this paper (along with any referred to as being attached or enclosed) is being hand delivered to Group Art Unit 1634 on the date shown below, addressed to the Commissioner for Patents, Reissue Litigation Box 7, Washington, D.C. 20231

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Date of Delivery

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U.S. Patent No. 4,554,088 (Whitehead *et al.*) discloses the use of single-stranded nucleic acid bound to dispersible magnetic beads to isolate complementary nucleic acid from a sample in a ligand-ligate binding reaction (*e.g.*, see column 17, lines 15-57).

U.S. Patent No. 4,563,419 (Ranki *et al.*) discloses a hybridization assay in which a target nucleic acid is (1) separated from other sample components by hybridizing the target to a complementary nucleic acid fragment affixed onto a nitrocellulose filter and (2) detected with a labeled probe.

U.S. Patent No. 4,672,040 (Josephson) discloses isolation of specific DNA or RNA fragments from a mixture of nucleic acid fragments, including the desired species, by using immobilization of a known probe to magnetic particles and placing the coupled particles in contact with the mixture to allow hybridization, followed by magnetic separation of the particles from unbound materials, and washing of the hybridized fragments on the magnetic particles (*e.g.*, see column 17, line 48 to column 19, line 10).

U.S. Patent No. 4,683,202 (Mullis) discloses the polymerase chain reaction (PCR) method of nucleic acid amplification.

EPO Publication No. 0 328 829 is the published (Aug. 23, 1989) European patent application that corresponds to U.S. application no. 07/136,920, that is substantially identical to the disclosure of the present reissue application.

Arsenyan *et al.*, *Gene* 11:97-108 (1980) discloses the isolation and amplification of rat liver 5S RNA genes that relies on preliminary enrichment of the genes, followed by amplification by bacterial cloning. For preliminary enrichment of the 5S RNA genes, the individual (+) and (-) gene strands are captured from a sample containing denatured DNA fragments on solid supports with bound capture probes (oligo(dT) cellulose or 5S cDNA-cellulose). The (+) and (-) strands are separated from the sample, eluted from the solid supports, hybridized together, cloned into a vector, and amplified in *E. coli* transformants grown *in vitro*.

Blumenthal, *Proc. Natl. Acad. Sci. U.SA.* 77(5):2601-2605 (1980) discloses transcription of RNA species mediated by the enzyme Qß replicase under particular conditions.

Boss et al., *J. Biol. Chem.*, 256(24):12958-12961 (1981) discloses isolation of a target yeast iso-1-cytochrome c (CYC1) mRNA by hybridization to a complementary cloned DNA attached to a solid matrix (*e.g.*, diazobenzyloxymethyl cellulose powder), followed by sequencing using a CYC1-specific oligonucleotide primer and the dideoxy chain termination method. The sequencing reaction produces a multitude of sequences from the target nucleic acid, which are detected by gel separation and autoradiography.

Brown *et al.*, *Ann. Rev. Biochem.* 43:667-693 (1974) discloses methods of isolating nucleic acid sequences by using polynucleotides fixed to insoluble matrices, and the desirability of combining nucleic acid isolation with subsequent amplification. Brown et al., at pages 673-674, discloses DNA purification by using an affinity column in which complementary RNA or DNA molecules are fixed to an insoluble support (*e.g.*, nitrocellulose or cellulose) and circulating the soluble DNA mixture through the affinity column. Brown et al., at page 687, paragraph 2, states that "purification of important structural genes will have to be coupled with some method in which a small amount of a given gene can be increased enormously in amount. After purification has enriched the gene sequence ... the remaining DNA would be amplified hundreds to thousandsfold in amount.... The amplification step might be carried out in vitro by an efficient DNA polymerase, which would replicate faithfully each molecule of DNA many times."

Burgess, "Purification and Physical Properties of *E. coli* RNA Polymerase" *in* RNA Polymerase (Losick and Chamberlin, eds.) (Cold Spring Harbor Laboratory, Cold Spring Harbor, NY, 1976), part 1: 69-100, discloses procedures for purifying RNA polymerases and determining enzyme purity, commonly associated enzymatic contaminants (pages 86-89) and properties of RNA polymerases.

Feinberg *et al., Anal. Biochem.* 132:6-13 (1983) discloses a radiolabeling method that uses random hexamer oligonucleotide primers to initiate non-specific enzymatic reproduction of isolated polynucleotides.

Gaubatz *et al., Biochim. Biophys. Acta,* 825:175-187 (1985) discloses a method of cDNA strand displacement synthesis to amplify mRNA sequences.

Kornberg, DNA Synthesis (W.H. Freeman & Co., San Francisco, CA, 1974), page 65 ("Substrates"), discloses the effects of enzymatic contaminants on DNA polymerase mediated reactions, such as an endonuclease that may "convert an active template-primer into an inhibitor that binds the enzyme in an unproductive complex", an exonuclease that "can enlarge nicks into gaps" or "an excess of nuclease [that] would lead to net loss of DNA."

Maniatis et al., Molecular Cloning, A Laboratory Manual (Cold Spring Harbor Laboratory, Cold Spring Harbor, NY 1982), Chapter 6, pages 187-196, discloses methods of isolating RNA that include inhibiting RNases or inactivating nucleases, and selecting poly(A)+ RNA using oligo(dT)-cellulose. It emphasizes the need to use nuclease-free laboratory ware and to carefully prepare solutions to avoid contamination.

Maniatis et al., Molecular Cloning, A Laboratory Manual (Cold Spring Harbor Laboratory, Cold Spring Harbor, NY 1982), Chapter 7, pages 213-214, discloses synthesis of first strand cDNA using reverse transcriptase, emphasizing problems associated with contaminating RNase in the reaction.

McGraw-Hill's *Biotechnology Newswatch* 6(19): 8 (Oct. 6, 1986), "DNA cleavage adapter groomed for genetic diagnostics" at paragraph 7, quotes K. Mullis, inventor of the polymerase chain reaction (PCR), stating that his technique results in "a lot of other things replicating that you don't want." The article describes a combination of PCR with other techniques to produce an improved diagnostic assay.

Orkin, *N. Engl. J. Med.*, 317(16):1023-1025 (1987) discloses the use of elevated temperature and heat-stable polymerase in PCR to minimize the problem of background amplification due to cross-hybridization of primers to non-target sequences at a lower temperature (see page 1024, column 2).

Polsky-Cynkin *et al., Clin. Chem.*, 31(9):1438-1443(1985) discloses sandwich hybridization assays in which a target DNA is captured by a complementary probe affixed to a solid support (beads, polypropylene test tubes or polypropylene solid-phase receptacles), and detected on the solid support by using a radiolabeled probe.

Powell *et al., Cell,* 50:831-840 (1987) discloses capture of poly(A)$^+$ RNA from a sample by using oligo(dT)-cellulose chromatography, followed by PCR amplification of cDNA made from the eluted RNA and detection of the amplified products by using radioactively labeled oligonucleotides (see Experimental Procedures on pages 838-839).

Saiki *et al.*, *Nature*, 324: 163-166 (1986), discloses that reduced signals in a assay based on PCR amplification may result from failure to purify target DNA, *i.e.*, "inhibition of the amplification process by cellular debris" (see text spanning page 164, column 2 to page 165, column 1).

Syvänen *et al., Nuc. Acids Res.*, 14(12):5037-5048 (1986) discloses a hybridization assay in which a target nucleic acid is captured by hybridization to a capture probe with an affinity label (*e.g.*, biotin) and then binds to an affinity matrix (e.g., streptavidin agarose beads) through an affinity interaction (*e.g.*, biotin-avidin interaction). The capture probe and target nucleic acid are hybridized in solution to take advantage of solution-phase kinetics (see pages 5042-5043).

Thompson *et al., Clin. Chem.*, 35(9): 1878-1881 (1989) discloses a hybridization assay that combines reversible target capture, essentially as disclosed in the present reissue application, with enzymatic amplification (PCR) of the purified target nucleic acid.

This information has been served on applicant in accordance with 37 C.F.R. 1.248, as indicated by the attached proof of service.

Respectfully submitted,

Date: August 1, 2000

By: Peter R. Shearer

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Registration No. 28,117

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FORM PTO-1449 U.S. DEPARTMENT OF COMMERCE PATENT AND TRADEMARK OFFICE	GPI ATTY. DOCKET NO.	SERIAL NO. / PATENT NO.
	Collins - Reissue Protest	09/533,906 / 5,570,338
INFORMATION DISCLOSURE STATEMENT BY APPLICANT	APPLICANT	
	Collins *et al.*	
(USE SEVERAL SHEETS IF NECESSARY)	REISSUE FILING DATE	GROUP
	March 8, 2000	1634

U.S. PATENT DOCUMENTS

EXAMINER INITIAL		DOCUMENT NUMBER	DATE	NAME	CLASS	SUBCLASS	FILING DATE (IF APPROPRIATE)
		4 5 5 4 0 8 8	11/19/85	Whitehead *et al.*	252	62.54	
		4 5 6 3 4 1 9	01/07/86	Ranki *et al.*	435	6	
		4 6 7 2 0 4 0	06/09/87	Josephson	436	526	
		4 6 8 3 2 0 2	07/28/87	Mullis	435	91	

FOREIGN PATENT DOCUMENTS

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		0 3 2 8 8 2 9	08/23/89	EPO				

EXAMINER INITIAL		**OTHER DOCUMENTS** (INCLUDING AUTHOR, TITLE, DATE, PERTINENT PAGES, ETC.)
		Arsenyan *et al.*, "Isolation of rat liver 5S RNA genes", *Gene*, 11:97-108 (1980)
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		Boss *et al.*, "Sequence of the Yeast Iso-1-Cytochrome *c* mRNA", *J. Biol. Chem.*, 256(24):12958-12961 (1981)
		Brown *et al.*, "Methods of Gene Isolation", *Ann. Rev. Biochem*, 43:667-693 (1974)
		Burgess, "Purification and Physical Properties of *E. coli* RNA Polymerase", *in* RNA Polymerase (Losick and Chamberlin ed.), (Cold Spring Harbor Laboratory, Cold Spring Harbor, NY, 1976), pt. 1:69-100
		Feinberg *et al.*, "A Technique for Radiolabeling DNA Restriction Endonuclease Fragments to High Specific Activity", *Anal. Biochem.*, 132:6-13 (1983)
		Gaubatz *et al.*, "Displacement synthesis of globin complementary DNA: evidence for sequence amplification", *Biochim. Biophys. Acta*, 825:175-187 (1985)
		Kornberg, DNA Synthesis, (W.H. Freeman and Co., San Francisco, CA, 1974), pg. 65

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		Polsky-Cynkin *et al.*, "Use of DNA Immobilized on Plastic and Agarose Supports to Detect DNA by Sandwich Hybridization", *Clin. Chem.*, 31(9):1438-1443 (1985)
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		Saiki *et al.*, "Analysis of enzymatically amplified β-globin and HLA-DQα DNA with allele-specific oligonucleotide probes", *Nature*, 324:163-166 (1986)
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References cited by Protester

THE UNITED STATES PATENT AND TRADEMARK OFFICE

In re Reissue Application of: U.S. Patent No. 5,750,338 Mark L. Collins *et al.* Reissue Serial No. 09/533,906 Reissue Application Filed: March 8, 2000 For: TARGET AND BACKGROUND CAPTURE METHODS WITH AMPLIFICATION FOR AFFINITY ASSAYS	Group Art Unit: 1634 Examiner: Unassigned

PROOF OF SERVICE

ATTENTION: REISSUE LITIGATION BOX 7
Assistant Commissioner for Patents
Washington, D.C. 20231

I, Kellee Clinton, do hereby declare as follows:

1. I am over the age of eighteen years, and not a party to this proceeding.

2. My business address is 10210 Genetic Center Drive, San Diego, California 92121.

3. On August 1, 2000, I served the following documents in the manner described in Paragraph 4:

PROTEST UNDER 37 C.F.R. § 1.291;

ATTACHMENT A TO PROTEST ('338 PATENT HISTORY...);

DECLARATION OF MICHAEL M. HARPOLD, Ph.D;

EXHIBIT A – CURRICULUM VITAE OF MICHAEL M. HARPOLD, Ph.D;

INFORMATION DISCLOSURE STATEMENT ACCOMPANYING PROTEST;

PTO FORM 1449;

TWENTY-ONE (21) REFERENCES;

TRANSMITTAL LETTER TO PTO; and

COPY OF RETURN/PREPAID POSTCARD TO PTO.

4. On August 1, 2000, I served the documents listed in Paragraph 3 by placing a copy of each of the documents in an envelope, sealing the envelope, and, with postage fully prepaid, placing the envelope for deposit with the United States Postal Service on the same day, at my business address shown above, following ordinary business practices, addressed to:

Jean Burke Fordis, Reg. No. 32,984
Finnegan, Henderson, Farabow, Garrett & Dunner, L.L.P
Suite 700
1300 I Street, N.W.
Washington, D.C. 20005-3315

I hereby declare under the laws of the United States of America that all statements made herein are true of my own knowledge and further that these statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment or both, under Section 1001 of Title 18 of the United States Code.

Executed at San Diego, California on August 1, 2000.

Kellee Clinton

FORM PTO-1449 U.S. DEPARTMENT OF COMMERCE PATENT AND TRADEMARK OFFICE	GPI ATTY. DOCKET NO. Collins - Reissue Protest	SERIAL NO. / PATENT NO. 09/533,906 / 5,570,338
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